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THE ADRENAL MEDULLA IN VARIOUS DISEASES

A HISTOPHYSIOLOGIC STUDY

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In 1928 Elaut [1] described the adrenal medulla from the points of view of histophysiology and histopathology. He studied structural and cytologic changes in the medullas of experimental animals after injection of strychnine, insulin and bacterial toxins, and he described the effects of shock, infection and hemorrhage on the human adrenal gland. From these observations he concluded that the function of the medulla of the adrenal gland can be evaluated with a high grade of certainty from a cytologic and histologic study of well preserved organs. He observed that four signs characterize increased function of the medulla: (1) dilatation of the sinusoids with swelling of their endothelium; (2) dilatation of the intratrabecular and intercellular secretory canals; (3) presence of vacuoles in the medullary cells and of secretary globules in the canals and (4) enlargement of cells and nuclei with formation of giant cells.

While Elaut's observations have been confirmed by Goormaghtigh [2] in a study of animals exposed to low temperatures, no studies have appeared in the literature on the relationship of structure and function of the adrenal medulla in man.

The present paper is based on a cytologic and histologic study of the medulla of the adrenal gland in 125 cases in which autopsy was done. The purpose of the investigation was to see whether there was any correlation between the structure and the function of the medulla. Especially were we interested in this problem as it might aid in deciding the possible role which the medulla plays in essential hypertension.

METHOD

The adrenal glands were obtained at autopsies in St. Francis Hospital. After fixation in 4 per cent formaldehyde solution, they were cut into fine parallel sections with a razor, and the slices showing the widest extension of the medullary portion were embedded in paraffin. The cytologic and histologic observations on sections stained with hematoxylin and eosin were entered in a list without knowledge of the clinical or the pathologic diagnosis in any case. While Elaut did not measure the cells and nuclei, we decided to obtain objective values. The nuclei and cells were measured in every case in blocks of 100, from 100 different areas, chosen at random according to where the scale of the micrometer eye piece happened to fall as the slide was systematically moved across the stage.

THE NORMAL MEDULLA

Most textbooks describe the cells of the medulla of the adrenal gland as being irregularly arranged in cords and whorls and make no mention of any orderly arrangement of cells. If one examines a gland fixed by perfusion—and most of our material was obtained from bodies previously preserved by arterial embalming—one finds that the blood vessels are not collapsed but are open and free of blood, fixed in distention; the relationship of cells to vessels in such sections is much clarified. We found that around the veins the medullary cells were arranged as columnar epithelium and that the "whorl" described in unperfused glands is in reality the cells surrounding a collapsed vessel cut in cross section whereas the "cord" is cells similarly arranged cut parallel to the long axis of the vein. Every medullary cell comes to touch or to lie close to one of the veins or its branches. As a rule, the cords or trabeculae are composed of a double row of medullary cells. In the axis of each trabecula runs a small canal, supported here and there by extremely fine reticulum fibrils. These canals, first described as preformed structures by Félicine,[3] communicate on one hand with intercellular spaces and on the other hand with blood vessels. The communications with

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1. Elaut, E.: Arch. internat. de méd. exper. **6**:69, 1928.
2. Goormaghtigh, N.: Arch. de biol. **41**:109, 1931.
3. Félicine, L.: Arch. f. mikr. Anat. **63**:283, 1903-1904.

the latter are so narrow that normal passage of red blood cells is not possible. The cells are arranged perpendicularly to the wall of the vein. The cell nucleus is situated, as a rule, in the pole opposite the implantation, adjacent to the intra-

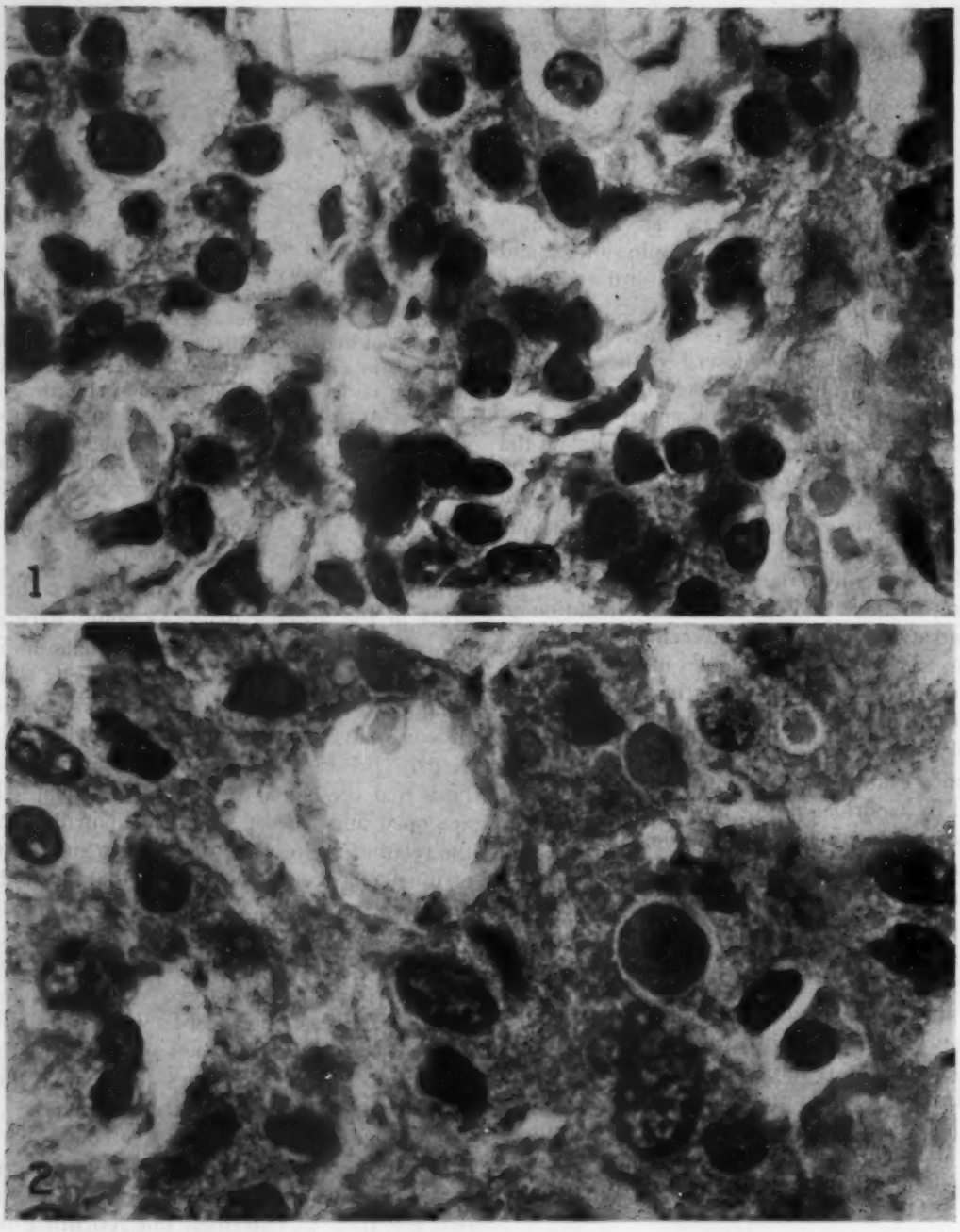

Fig. 1.—Adrenal medulla of low activity. Note that cells and nuclei are small. × 1,200.
Fig. 2.—Adrenal medulla of high activity. The cells and nuclei are large. Acidophilic globules are present in secretory canals. There are giant nuclei. × 1,200.

trabecular canal. This is best demonstrated in cross sections of trabeculae.

Elaut distinguished two types of medullary cells: (1) small, narrow cells, strongly chromaffin and osmophilic with granular cytoplasm, and (2) large cells with clear cytoplasm containing vacuoles and a large nucleus. Between these two cell types all stages of transition occur, and apparently all cell forms represent different functional phases of the medullary cell. There are, according to Elaut and Goormaghtigh, zones of different functional activity in the same gland. In man, horse and dog the more active zones,

characterized by large clear cells, are found in the periphery of the medulla adjacent to the cortex, where the medulla receives its blood supply.

HISTOPHYSIOLOGIC OBSERVATIONS

Table 1 summarizes the histophysiologic observations on our material. According to Elaut's criteria of increased function of the medulla, the 125 cases were divided into three groups: 38 cases in which activity was low, 51 in which it was moderate and 36 in which it was high.

Age.—In the first decade of life as a rule activity of the medulla is low (table 2). This observation is in accord with the view of physiologists that the blood pressure is relatively low until the onset of puberty (Best and Taylor [4]). In no case in which the patient was under 15 years of age did the medulla show high activity. There were 3 cases in which activity was moderate during childhood. The glands were from male twins 1 year of age, both of whom died from bronchopneumonia, and an 8 year old boy with streptococcic septicemia. Of the cases in which the patients were adults, activity of the medulla was low in 22, moderate in 43 and high in 34. While the average ages in these three functional groups were about the same—48, 52 and 54 years, respectively—increased activity was more frequent at the higher ages. In the first group, with low activity, the ratio of patients over 50 years to those under 50 was approximately 1 to 2; in the second, 1 to 1, and in the group with high activity, 2 to 1.

Sex.—In 81 of our cases the patients were males, and in 44, females. While in our first group the ratio of males to females was about 2 to 1, we found almost three times as many males as females among the cases in which there was increased function of the medulla.

Infectious Diseases.—In 35 cases active inflammatory disease was the cause of death. Among the 35 patients were 19 adults and 16 children and adolescents. Of the adults, 15 had acute inflammation, 3 had septicemia from various portals, 5 had pneumonia or bronchopneumonia, 2 peritonitis and 1 pericarditis.

There were 4 cases in which chronic inflammatory diseases were noted, namely, subacute endocarditis, abscesses in the liver and in the subphrenic space and bronchiectasis.

High activity of the medulla was evident in only 2 adults with inflammatory disease—one with streptococcic septicemia of a fulminating type and the other with endocarditis; in 9 adults moderate activity and in 8 inactivity of the medulla was noted.

In the young age group 9 patients died from acute inflammatory disease. Six of these had low activity and 3 moderately high activity of the medulla. The histologic observations in cases in which the diagnoses were rheumatic carditis, diphtheria, encephalitis and poliomyelitis varied.

In surveying these cases it becomes apparent that chronic inflammatory diseases are associated with low activity of the medulla and that the changes in medullary activity in acute infections are inconsistent. No definite correlation was noted between the structural changes in the medulla and the type of infecting organism.

Neoplastic Diseases.—Malignant neoplastic diseases were present in 11 cases. If we disregard 1 case in which carcinoma of the prostate was observed associated with high blood pressure there were no cases in which cancer was accompanied with high activity of the adrenal medulla. In 5 cases the medulla showed low function and in 5 others moderate activity. Nor was the morphologic picture of the medulla influenced by the location, the size or the rapidity of growth of the tumor.

Trauma and Poison.—There were 12 cases in which death was due to unnatural causes. Of medullas from adults, only 1 exhibited low activity. It was from a 40 year old Negro woman with multiple stab wounds in the abdomen. In the others activity was either moderate or increased. We are able to confirm Elaut's statement that increased activity of the medulla is definitely associated with injuries of the brain. The only young person in this group, a 15 year old girl, who died from extensive burns, had high medullary activity. Among the persons whose death was due to poison, a 39 year old woman who had committed suicide by taking a large dose of strychnine had high activity of the medulla. Another woman who had inhaled naphtha fumes had only a moderately active medulla. The only inactive medulla among the adolescent persons of this group was that of a 19 year old youth who had died during anesthesia induced with ether.

Pregnancy.—In 3 cases pregnancy was observed. A 39 year old Mexican woman died during childbirth from rupture of the uterus causing acute anemia. The medulla showed moderate activity. Hemorrhagic shock was also the cause of death in the second case. The patient was a 23 year old white woman who died from rupture of an ovarian tube in tubal pregnancy. The medulla revealed exceptionally high activity. The third case was complicated by

4. Best, C. H., and Taylor, N. B.: The Physiological Basis of Medical Practice, Baltimore, Williams & Wilkins Company, 1939, p. 208.

TABLE 1.—*Summary of Histophysiologic Observations*

Case	Age of Patient	Sex	Disease	Heart Weight, Gm.	Veins	Canals	Diameters of Cells and Nuclei, Microns	
							Cells	Nuclei
			1. Low Activity of the Medulla					
1	3 days	F	Atresia of aortic valve	50	Wide	Wide	9.5	6.3
2	5 days	M	Coarctation of aorta	40	Wide	Narrow	5.4	4.4
3	8 days	M	Erythroblastosis	50	Narrow	Narrow	8.4	7.1
4	2 wk.	F	Interstitial pneumonia	45	Narrow	Narrow	7.5	5.7
5	1 mo.	F	Transposition of great vessels	40	Narrow	Narrow	8.2	6.4
6	3 mo.	F	Bronchopneumonia	45	Wide	Narrow	9.5	5.7
7	4 mo.	M	Interstitial pneumonia	40	Medium	Medium	8.9	6.1
8	5 mo.	F	Interstitial pneumonia	45	Wide	Wide	8.7	6.4
9	6 mo.	F	Rickets, bronchopneumonia	38	Narrow	Narrow	12.0	7.6
10	5 yr.	M	Tetanus	140	Medium	Medium	11.0	7.7
11	8 yr.	F	Diphtheria	120	Narrow	Narrow	10.5	6.8
12	10 yr.	F	Rheumatic carditis	400	Wide	Wide	10.7	8.1
13	17 yr.	F	Encephalitis	240	Medium	Narrow	12.0	8.6
14	19 yr.	M	Death from anesthesia	280	Wide	Wide	10.8	6.1
15	2½ yr.	M	Diphtheria	60	Narrow	Narrow	11.6	8.1
16	4 yr.	M	Bronchopneumonia	80	Wide	Wide	8.9	6.7
17	24 yr.	F	Lymphoblastic leukemia	220	Medium	Medium	11.4	7.1
18	33 yr.	M	Gastric ulcer, bronchopneumonia	310	Narrow	Narrow	10.8	6.3
19	37 yr.	M	Mesenteric sarcoma	210	Wide	Narrow	6.5	5.3
20	37 yr.	M	Acute porphyria	350	Wide	Wide	11.2	7.8
21	38 yr.	F	Cirrhosis of liver	270	Medium	Medium	8.4	6.8
22	38 yr.	M	Polycystic kidneys	380	Medium	Medium	12.0	6.8
23	38 yr.	F	Pellagra	210	Wide	Medium	11.4	7.0
24	40 yr.	F	Abdominal stab wound	250	Medium	Medium	10.5	6.8
25	42 yr.	M	Hypernephroma	450	Medium	Narrow	8.2	5.5
26	43 yr.	M	Acute septicemia	400	Wide	Wide	11.8	6.8
27	43 yr.	M	Abscesses of liver	280	Narrow	Medium	11.9	8.2
28	46 yr.	M	Coronary occlusion	380	Wide	Wide	11.4	7.6
29	49 yr.	M	Postoperative peritonitis	350	Medium	Medium	11.4	6.8
30	53 yr.	F	Cancer of lung	240	Narrow	Narrow	9.5	6.5
31	55 yr.	F	Bronchiectasis	230	Medium	Medium	12.0	6.6
32	50 yr.	M	Subphrenic abscess	300	Wide	Wide	10.6	8.4
33	57 yr.	F	Mesenteric thrombosis, diabetes	350	Narrow	Narrow	11.8	7.0
34	59 yr.	M	Cancer of lung	450	Medium	Medium	7.5	5.4
35	62 yr.	M	Syphilitic aneurysm	400	Wide	Wide	11.6	8.6
36	62 yr.	M	Coronary occlusion	500	Wide	Wide	10.5	6.3
37	71 yr.	M	Peritonitis	360	Medium	Medium	11.8	8.0
38	81 yr.	F	Acute cholecystitis	350	Narrow	Narrow	9.4	7.3
			2. Moderate Activity of the Medulla					
1	1 yr.	M	Bronchopneumonia	70	Medium	Medium	13.7	8.4
2	1 yr.	M	Bronchopneumonia	44	Medium	Medium	13.8	8.4
3	8 yr.	M	Streptococcic sepsis	160	Narrow	Narrow	13.3	7.4
4	11 yr.	F	Diabetes, bronchopneumonia	180	Medium	Narrow	13.9	8.6
5	12 yr.	M	Rupture of aneurysm of cerebral artery	230	Wide	Narrow	12.7	7.1
6	17 yr.	M	Poliomyelitis	280	Medium	Medium	13.1	7.0
7	14 yr.	M	Rheumatic carditis	610	Wide	Wide	12.2	10.8
8	20 yr.	M	Gunshot wound through heart	300	Medium	Narrow	14.4	10.3
9	26 yr.	F	Eclampsia	380	Medium	Medium	13.9	9.9
10	31 yr.	F	Naphtha poisoning	200	Medium	Narrow	13.7	7.5
11	31 yr.	F	Burns	350	Wide	Wide	13.1	8.6
12	32 yr.	M	Basal fracture	330	Medium	Medium	14.6	7.8
13	35 yr.	M	Gunshot wound of brain	300	Narrow	Narrow	14.4	8.4
14	38 yr.	M	Fracture of skull	340	Wide	Wide	12.2	6.8
15	38 yr.	F	Coronary occlusion	280	Wide	Wide	12.8	7.7
16	39 yr.	F	Rupture of uterus	300	Medium	Medium	13.9	8.9
17	39 yr.	F	Strychnine poisoning	220	Medium	Medium	14.3	9.9
18	41 yr.	M	Intestinal obstruction	350	Wide	Wide	12.7	8.1
19	42 yr.	F	Cancer of lung	200	Wide	Medium	13.4	8.6
20	44 yr.	M	Rib fractures	350	Medium	Medium	13.3	6.2
21	42 yr.	F	Cancer of rectum metastatic to brain	270	Wide	Wide	14.6	8.6
22	42 yr.	M	Tubular nephritis	620	Narrow	Narrow	14.3	6.0
23	45 yr.	M	Aortic stenosis	700	Narrow	Medium	13.3	8.5
24	45 yr.	M	Paralytic ileus, acromegaly	450	Medium	Wide	12.9	8.0
25	45 yr.	M	Coronary occlusion, hypertension	410	Narrow	Wide	14.8	8.7
26	45 yr.	M	Chronic glomerulonephritis	600	Narrow	Narrow	13.3	7.6
27	47 yr.	M	Cirrhosis of liver	310	Wide	Wide	14.8	8.8
28	47 yr.	F	Cancer of mouth, bronchopneumonia	250	Medium	Narrow	12.7	9.5
29	49 yr.	M	Gunshot wound of brain	200	Medium	Medium	13.0	8.6
30	53 yr.	M	Cerebral hemorrhage	300	Wide	Narrow	14.6	8.4
31	53 yr.	M	Postoperative bronchopneumonia	310	Wide	Wide	14.2	7.8
32	54 yr.	M	Abdominal gunshot wound	500	Medium	Medium	13.3	9.1
33	55 yr.	M	Coronary occlusion	380	Medium	Wide	14.6	9.5
34	56 yr.	M	Rupture of syphilitic aneurysm	290	Medium	Medium	14.6	8.4
35	57 yr.	F	Pellagra	330	Medium	Medium	13.3	7.6
36	57 yr.	F	Macrocytic anemia	180	Narrow	Narrow	14.8	9.2
37	58 yr.	M	Mesenteric sarcoma	350	Medium	Medium	14.4	8.7
38	58 yr.	M	Pulmonary embolism	400	Medium	Medium	14.8	8.4
39	58 yr.	F	Mesenteric thrombosis	260	Medium	Medium	13.3	7.6
40	58 yr.	M	Bronchopneumonia	450	Medium	Medium	13.5	7.6
41	61 yr.	M	Pyelonephritis	410	Narrow	Medium	13.2	8.4
42	63 yr.	M	Tumor of brain	360	Medium	Medium	13.1	8.7
43	66 yr.	M	Ulcerative endocarditis	350	Wide	Wide	13.7	8.3
44	64 yr.	F	Peritonitis following appendicitis	200	Wide	Medium	14.4	8.9
45	68 yr.	F	Cerebral hemorrhage	430	Medium	Medium	14.1	8.7
46	63 yr.	M	Hypertrophy of prostate, pyelonephritis	600	Wide	Medium	12.4	9.6
47	73 yr.	M	Myocardial infarct	530	Medium	Medium	14.8	8.6
48	73 yr.	M	Lobar pneumonia	400	Wide	Wide	13.7	8.6
49	74 yr.	M	Empyema, pericarditis	500	Wide	Wide	12.4	8.2
50	74 yr.	F	Lymphatic leukemia	260	Wide	Wide	14.6	8.6
51	82 yr.	M	Nephrosclerosis	530	Wide	Wide	14.3	9.8

TABLE 1.—*Summary of Histophysiologic Observations.—Continued*

Case	Age of Patient	Sex	Disease	Heart Weight, Gm.	Veins	Canals	Diameters of Cells and Nuclei, Microns	
			3. High Activity of the Medulla				Cells	Nuclei
1	15 yr.	F	Burns	250	Wide	Medium	15.4	10.1
2	16 yr.	F	Chronic glomerulonephritis	430	Medium	Medium	16.1	8.7
3	22 yr.	M	Subacute endocarditis	460	Medium	Medium	15.1	7.9
4	23 yr.	F	Ruptured tubal pregnancy	250	Narrow	Narrow	16.5	8.6
5	28 yr.	F	Chronic glomerulonephritis	300	Medium	Medium	15.6	9.5
6	29 yr.	M	Bronchopneumonia, hypertension	300	Wide	Wide	16.8	8.4
7	32 yr.	M	Malignant hypertension	800	Medium	Medium	17.3	9.5
8	37 yr.	F	Obesity, syphilis	310	Wide	Narrow	15.4	8.6
9	41 yr.	M	Hypertension	660	Wide	Medium	15.4	8.2
10	43 yr.	M	Pneumonia	430	Narrow	Narrow	15.8	8.7
11	44 yr.	M	Prostatic carcinoma	500	Narrow	Narrow	15.2	9.1
12	45 yr.	M	Hypertension	610	Narrow	Narrow	16.5	9.5
13	47 yr.	M	Bronchopneumonia	500	Medium	Medium	16.2	9.7
14	47 yr.	M	Subarachnoid hemorrhage	450	Medium	Wide	15.2	9.7
15	51 yr.	M	Coronary occlusion	370	Wide	Wide	15.8	8.7
16	53 yr.	F	Thyrotoxic crisis	280	Medium	Medium	15.8	8.4
17	53 yr.	M	Rupture of heart	650	Medium	Medium	16.9	9.1
18	54 yr.	F	Myocardial infarct	440	Wide	Medium	16.0	8.9
19	55 yr.	F	Subarachnoid hemorrhage	455	Medium	Medium	16.5	9.5
20	58 yr.	M	Toxic goiter, cancer of lung	630	Medium	Narrow	16.3	9.6
21	58 yr.	M	Hypertension	600	Medium	Wide	17.0	9.5
22	59 yr.	M	Cancer of colon, obesity	420	Medium	Narrow	15.9	8.9
23	59 yr.	M	Myocardial infarct	480	Medium	Medium	15.4	8.8
24	60 yr.	M	Myocardial infarct	710	Narrow	Narrow	19.0	9.7
25	60 yr.	M	Cerebral thrombosis	550	Wide	Medium	15.5	9.2
26	62 yr.	F	Hypertension	710	Medium	Medium	16.0	10.2
27	65 yr.	M	Hypertension	760	Wide	Wide	15.5	9.0
28	68 yr.	F	Diabetes, hypertension	450	Medium	Narrow	16.3	9.9
29	66 yr.	M	Streptococcic sepsis	500	Medium	Medium	15.3	8.9
30	66 yr.	M	Myocardial infarct	636	Medium	Medium	16.5	9.1
31	72 yr.	M	Hypertension	720	Wide	Wide	19.8	8.4
32	72 yr.	M	Cerebral hemorrhage	490	Wide	Wide	15.2	8.7
33	72 yr.	F	Cerebral thrombosis	350	Wide	Narrow	15.6	9.1
34	74 yr.	M	Rupture of heart	450	Narrow	Narrow	15.5	7.9
35	76 yr.	M	Coronary occlusion	550	Wide	Narrow	15.2	8.2
36	85 yr.	M	Diabetes, hypertension	400	Medium	Medium	15.2	9.5

eclampsia after delivery of stillborn twins. The medulla showed signs of moderate activity. If any conclusions can be drawn from these few cases, it would appear that acute hemorrhagic shock does not stimulate the function of the medulla and that pregnancy is associated with high medullary activity only in the early months.

Endocrine and Metabolic Diseases.—In the 14 cases belonging to this group the histophysiologic changes in the adrenal glands were difficult to evaluate because in many instances the metabolic disorders were associated with infectious, neoplastic or vascular diseases.

TABLE 2.—*Relationship Between Age and Activity of the Medulla*

Age	Low Activity	Moderate Activity	High Activity
1 to 10 yr.	14	3	..
11 to 20 yr.	2	5	2
21 to 30 yr.	1	1	4
31 to 40 yr.	7	8	2
41 to 50 yr.	6	12	6
51 to 60 yr.	4	11	10
61 to 70 yr.	2	6	6
71 to 80 yr.	1	4	5
81 to 90 yr.	1	1	1

The medulla of a 6 month old girl with severe rickets, undernourishment and terminal bronchopneumonia appeared to be inactive. The medullas from 2 middle-aged women with pellagra showed low activity and that from a 38 year old white man with acute porphyria also showed low function. The medulla of a 45 year old man of the acromegalic type who died of paralytic ileus showed moderate activity.

Hyperfunction of the medulla was observed in 7 cases of this group. In 2 of these cases obesity was prominent. One of these was complicated with syphilitic aortitis; the other, with perforating carcinoma of the colon. There were 2 cases in which thyrotoxicosis was present. One of the patients died of thyrotoxic crisis after operation; the other, of a large bronchogenic carcinoma. In the other 3 cases in which high activity of the medulla was observed diabetes was a feature. Two of the diabetic patients were adults and the third was a young girl who died in diabetic coma.

Inflammatory and Neoplastic Diseases of the Urinary Organs.—There were 2 cases of renal tumor. One of the patients was a 38 year old man with bilateral polycystic kidney who died in uremic coma, and the other was a 42 year old man with hypernephroma. Both showed low function of the adrenal medulla.

Hypertrophy of the prostate in an elderly man and carcinoma of the prostate in a 44 year old man had resulted in severe pyelonephritis and were associated with moderate activity of the medulla.

In 5 cases of chronic glomerulonephritis the microscopic appearance of the adrenal glands varied. Two of the patients were men in the fifth decade of life. The medulla from each of these presented moderate activity. Three of the

patients were under 30 years of age. The medulla from each of these showed hyperfunction.

Coronary Occlusion.—In 12 cases of coronary occlusion the results were not uniform. In 5 cases in which the heart weight exceeded 500 Gm. association with hypertension was noted. The medullas of all 5 patients, who were between 60 and 76 years of age, showed high activity. In 2 of the remaining 7 cases medullary activity was low, a fact which was not explained by the weight of the heart or by the age of the patients. In 3 other cases moderate activity was recorded and in 2 in which the heart weights varied from 370 to 480 Gm. there was hyperfunction of the medulla. Summarizing the results in this group, we may state that in three fourths of the cases the arterial condition was associated with medullary function exceeding the normal. The hearts of the patients with low medullary function were as a rule smaller than those of patients with increased activity of the medulla. In all the cases in which a diagnosis of hypertension was justified on clinical and anatomic grounds a high activity of the medulla was exhibited.

Cerebral Vascular Disease.—In 9 cases cerebral hemorrhage or thrombosis was the cause of death. Two of the patients, a 12 year old boy with a ruptured aneurysm of a cerebral artery and a 68 year old man with massive spontaneous cerebral hemorrhage, showed moderate activity of the medulla. All others exhibited markedly increased function of the medulla regardless of the source of the hemorrhage, its size or its location; also the patients with thrombosis of cerebral arteries had hyperfunction of the medulla. The ages of these patients were between 53 and 76 years and all except 3 were men. The weights of their hearts were, as a rule, above normal, ranging from 390 to 550 Gm.

Hypertension.—Of our 26 cases of hypertension, 9 have been mentioned previously—4 under "Coronary Occlusion," and 5 under "Cerebral Vascular Diseases." The ages of the 26 patients were between 32 and 82 years, the average age being 61 years. As to sex a great majority, 19, were men. The hearts weighed from 450 to 860 Gm.; the average weight was 587 Gm. Rupture of the heart was the immediate cause of death in 2 cases, while streptococcic septicemia caused death in 1 case and bronchopneumonia in 1.

In all but 2 cases the morphologic criteria of marked hyperfunction of the medulla were present. The diameters of the cell nuclei were between 8.2 and 10.2 microns; the diameters of the cells were between 15.2 and 19.8 microns. Giant nuclei were observed in varying number; they occurred frequently twelve times. In the majority of the cases secretory oxyphilic granules were present in the secretory canals, together with vacuoles in the cytoplasm of the medullary cells.

The 2 cases excepted, in which only moderate activity of the medulla was exhibited, were those of an 82 year old man whose heart weighed 530 Gm. and a 73 year old man with a myocardial infarct in a heart weighing 520 Gm.

COMMENT

From the study of the available material it has become apparent that distinct morphologic changes occur in the adrenal medulla in various systemic diseases. The functional interpretation of the cytologic and histologic alterations was based on the work of Elaut.

The most valuable criterion of activity of the medulla in our opinion is an increase in the size of the medullary cells and their nuclei associated with the presence of giant nuclei. The other signs of increased activity which Elaut pointed out—the dilatation of veins and secretory canals and the appearance of vacuoles—appear to be less reliable. We believe that the measurement of 100 cells and nuclei in a given medulla furnishes a practical index for judging its activity.

We observed during our histophysiologic studies of well preserved adrenal glands that during the first and second decade of life the activity of the medulla is low. In adults low function of the medulla was associated with chronic inflammatory diseases. We were unable to confirm the view held by Elaut that acute bacterial infections stimulate medullary function. In neoplastic diseases the activity of the medulla tended to be low, especially when there was cachexia. We agree with Elaut that injuries of the brain are often associated with hyperfunction of the medulla and that strychnine stimulates the activity of this tissue. Traumatic shock and hemorrhage in our experience had no effect on the structure of the medulla. In regard to metabolic disorders we observed that diabetes, obesity and thyrotoxicosis are associated with definitely increased function of the medulla.

Of greatest interest were our observations in cases of hypertension. The fact that medullary activity was increased in almost all of our cases of hypertension conflicts with the present tendency to disregard entirely hormonal factors in the genesis of essential hypertension. The results of our studies corroborate the view held by

Vaquez,[5] Schur and Wiesel,[6] Frei,[7] Goldzieher,[8] Westphal[9] and Duthoit[10] that the adrenal glands play an active and substantial role in many forms of arterial hypertension. In our opinion the following observations support strongly the hormonal theory of hypertension: 1. A number of medullary tumors have been described which were accompanied by transitory and permanent arterial hypertension. Their pathogenic significance in regard to hypertension has been evidenced by spectacular cures obtained by their removal. 2. Low blood pressure is a characteristic feature of insufficiency of adrenal tissues, as observed in Addison's disease.

In 1907 Schur and Wiesel[6] reported that they had always observed a connection between hypertension and hyperplasia of the adrenal medulla. Schmorl,[11] Goldzieher and Molnár[12] confirmed this; Cohen and Aschoff[13] discredited the theory of Schur and Wiesel, because they noted hypertrophy of the medulla in only 11 of 35 cases of hypertension.

In 1934 Frei[7] collected 723 autopsy cases in which the anatomic characteristics of high blood pressure were observed. In 80 per cent a definite enlargement of the adrenal medulla was present. Opsahl[14] studied the adrenal glands in 476 autopsy cases. In his material were 54 cases of hypertension. Two thirds of the cases in which high blood pressure was combined with nephrosclerosis the adrenal glands weighed more than 15 Gm. Opsahl[14] concluded that inadequate renal filtration is the cause of hypertrophy of the medulla and that the hyperfunction of the medulla compensates for the insufficient filtration of the glomeruli by raising the blood pressure. Westphal and Sievert[9] examined microscopically 10 adrenal glands in cases of hypertension and noted in 5 glands marked hypertrophy of the medulla and in 9 glands veins with thick muscle layers as described first by Goldzieher.[15]

Duthoit[10] described the structural changes in adrenal glands obtained at autopsy in 9 cases of high blood pressure. In 6 definite hyperplasia of the medulla was demonstrated.

The interpretation of the fact that the adrenal medulla is hypertrophic in hypertension is open to question. While Schur and Wiesel,[6] Goldzieher,[8] Duthoit[10] and Westphal and Sievert[9] attributed an important and primary role in the genesis of hypertension to the medulla, von Lacadou[16] explained the enlargement of the medulla as a compensatory process, secondary to hypertrophy of the heart. Studying the size of the medulla at 68 autopsies, he came to the conclusion that not only in hypertension but also in valvular heart disease and in pulmonary emphysema hyperplasia of the medulla accompanies enlargement of the heart. He observed a close relationship between the size of the heart and the size of the medulla regardless of the cause of the hypertrophy of the heart. He expressed the belief that chronic cardiac strain increases the demand of the organism for epinephrine. Opsahl, on the other hand, is of the opinion that the damaged filtration of the sclerotic kidney calls for increased secretion of epinephrine to compensate for the narrowing of the capillary bed in the kidney.

TABLE 3.—*Relationship Between Increased Weight of the Heart and Activity of the Medulla in Cases in Which Hypertension Was Not Present*

Case	Disease	Heart Weight, Gm.	Activity of Medulla
1	Hypernephroma	450	Low
2	Cancer of lung	450	Low
3	Paralytic ileus	450	Moderate
4	Bronchopneumonia	450	Moderate
5	Subacute endocarditis	460	High
6	Coronary occlusion	500	Low
7	Abdominal gunshot wound	500	Moderate
8	Empyema, pericarditis	500	Moderate
9	Cancer of prostate	500	High
10	Hypertrophy of prostate	600	Moderate
11	Tubular nephritis	620	Moderate
12	Toxic goiter, cancer of lung	630	High
13	Aortic stenosis	700	Moderate

Our own observations confirm neither of these two theories. From table 3 it becomes apparent that there is no definite relationship betweeen weight of the heart and activity of the medulla. Further, Opsahl's theory that hypertrophy of the medulla is the result of a disturbance of glomerular filtration is not substantiated by our observations. While it is true that in the majority of our cases of chronic glomerulonephritis histologic evidence of increased activity of the adrenal gland was seen, there were many cases in which hypertension without disturbed function of the kidney was associated with the same or more increased activity of the adrenal medulla. In our opinion the results of our studies justify

5. Vaquez, H.: Semaine méd. **24**:45, 1904.
6. Schur and Wiesel, cited by Opsahl.[14]
7. Frei, W.: Frankfurt. Ztschr. f. Path. **46**:523, 1934.
8. Goldzieher, M. A.: Endocrinology **16**:20, 1932.
9. Westphal, K., and Sievert, C.: Ztschr. f. klin. Med. **133**:310, 1938.
10. Duthoit, A.: Progrès méd., 1935, p. 769.
11. Schmorl, cited by Opsahl.[14]
12. Goldzieher, M. A., and Molnár: Wien. klin. Wchnschr. **1**:212, 1908.
13. Cohen and Aschoff, L.: Verhandl. d. deutsch. path. Gesellsch. **12**:131, 1908.
14. Opsahl, R.: Acta med. Scandinav., 1938, supp. 92.
15. Goldzieher, M. A., and Sherman, I.: Arch. Path. **5**:1, 1928.
16. von Lacadou, W.: Klin. Wchnschr. **14**:1529, 1935.

the view that the hyperfunction of the medulla plays a primary role in the genesis of hypertension.

Anatomic and experimental observations have established the fact that renal ischemia is an important factor in fixed hypertension. However, the renal theory of hypertension fails to explain the transitory elevation of blood pressure in normal persons and in the initial stages of essential hypertension. As Leriche[17] pointed out, it is well known that during nervous excitation and mental anxiety—for instance, that in surgeons during operation and that in singers during performance—the blood pressure rises physiologically only to return to normal shortly afterward. A renal mechanism certainly cannot explain this phenomenon. Transitory elevations of the blood pressure are better explained by hypersecretion of the adrenal medulla, a view based on Cannon's conception of an emergency function of the adrenal gland. It is noteworthy that the same situations which produce a physiologic transitory elevation of blood pressure will also aggravate essential hypertension. In our opinion the problem of hypertension is not solved by experiments which raise the blood pressure permanently. So far experimental pathologists have been unable to reproduce the early symptoms and the evolution of essential hypertension. Even if we accept the importance of renal ischemia in the genesis of hypertension, the question arises: What causes the narrowing of the blood flow in the kidney, or, in other words, what mechanism acts as the silver clamp of Goldblatt in the spontaneous disease in man? Is it a spasm of the renal vessels? If so, what is the cause of the spasm? Perhaps the adrenal theory of hypertension gives the answer to this question. Not only is it possible to produce hyaline changes in the renal arterioles of animals by injection of epinephrine, but in cases of human hypertension caused by tumors of the adrenal medulla, vascular changes have been described which are identical with those seen in cases of essential hypertension.

17. Leriche, R.: Presse méd. **46**:489, 1938.

SUMMARY AND CONCLUSIONS

In 125 cases in which autopsy was done the adrenal glands were studied in an effort to correlate medullary changes with systemic diseases.

The histophysiologic criteria of Elaut served as a guide to the evaluation of the activity of the medulla. The measurement of 100 cells and 100 nuclei by means of a micrometer eye piece gave the best information on the function of a given medulla.

It was observed that in neoplastic diseases and in long-standing infections the activity of the medulla was decreased. High activity was noticed in injuries of the brain, diabetes, obesity and thyrotoxicosis. Twenty-six cases of hypertension not associated with inflammation of the kidneys were included in our series. In all except 2 histologic evidence of hyperfunction of the medulla was present. The grade of activity did not depend on the weight of the heart or on the presence of renal insufficiency.

The results of our studies support the theory that in the early stages of essential hypertension hormonal factors play a substantial and primary role.

ENCEPHALITIS COMPLICATING VIRUS PNEUMONIA

REPORT OF A CASE WITH AUTOPSY

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Although signs and symptoms referable to the nervous system have been rather frequently noted in patients with virus pneumonia, and leukocytosis and lymphocytosis reported in the spinal fluid in 2 cases,[1] respectively, the present report is, as far as I know, the first on the brain in death from encephalitis complicating virus pneumonia.

REPORT OF CASE

The patient, a school teacher aged 58, had a "cold" for one week. She taught school until two days before hospitalization, when cough developed and her breathing became asthmatic. She was admitted to Dr. William Leaman's service at the Hospital of the Woman's Medical College. Her temperature was 101 F.; the pulse rate was 84 and the respiratory rate 30 per minute. The blood pressure was 190 systolic and 100 diastolic. She was an obese woman, cyanotic, with labored respirations, too ill to give a history. Her pupils were regular and reacted to light. Rales were heard throughout her lungs. A roentgenogram was characteristic of virus pneumonia.

Oxygen therapy was instituted, but three and a half hours after admission she became suddenly worse. Stimulation was of no avail, and she died seven and a half hours after admission.

Laboratory studies had shown leukocytosis (leukocyte count, 12,850, 92 per cent polymorphonuclears), which may have been terminal. The urine contained albumin, numerous granular casts and a few leukocytes and bacteria.

Autopsy (thirteen and one-half hours after death).—The body showed extreme cyanosis. There was edema of the feet and ankles. There was a hemorrhagic area, 2 by 3 cm., in the right pectoral muscle.

The trachea was extremely congested and contained a thick hemorrhagic purulent exudate.

The pleurae presented diffuse fibrous adhesions at the lower lobes of both lungs. The lungs met in the midline. Both showed emphysema, especially of the upper lobes. The lower lobe of the right lung was subcrepitant. The cut surface of the right lung retracted normally as regards the upper lobe while the middle and the lower lobe were more congested, retracted less and were firmer in consistency. The left lung on palpation contained a firm area the size of a walnut in the lower lobe near the hilus. On section this area was wedge shaped, well defined and slightly raised; it did not reach the pleural surface; it was dark red, firm but not friable, and somewhat moist. The surrounding lung tissue was congested. In both lungs, but especially in the left, the bronchial mucosa was extremely congested and the bronchi contained thick blood-stained mucus.

The pericardium contained a normal amount of fluid. The epicardium showed numerous petechial hemorrhages. The heart weighed 580 Gm. There was abundant subepicardial fat. The heart muscle was soft, slightly mottled and showed numerous small patches of fibrosis. Both ventricles showed hypertrophy and dilatation. The right ventricular wall measured 1 cm. in thickness; the left ventricular wall, 3 cm. The cusps of the mitral valve were thickened, and the orifice was somewhat narrowed (1½ fingers). Two recent vegetations, gray and soft, were seen on the posterior leaflet. The other valves were not remarkable.

The spleen weighed 280 Gm. and was semidiffluent.

The right kidney weighed 205 Gm.; the left, 204 Gm. Both were pale yellow and apparently enlarged. The capsules stripped with moderate difficulty. The cut surface showed a pale yellowish, swollen cortex.

The liver weighed 2.20 Gm. and was congested.

The pancreas was congested but otherwise normal in gross structure.

The gastrointestinal tract showed congestion only.

The meninges were greatly congested. The brain weighed 1,650 Gm. and was extremely congested. There were no other gross changes.

Microscopic Observations.—(*a*) Right Bronchus: The epithelium was largely desquamated. In some places, however, it consisted of small heaped-up cells. The normal columnar cells had all been shed, but here and there irregular epithelial cells projected singly or in groups from the denuded membrane. Careful examination with oil immersion revealed that the protoplasm of many of them contained sharply defined round bodies about 1 micron in diameter. The bodies usually stained with eosin, although some were of a nondescript plum color. When the cells in which they occurred were not too degenerate, these bodies were seen to be surrounded by a halo. Occasionally several bodies were found in one cell. The bodies resembled those figured by Adams.[2] In the cell debris in the lumen masses of similar bodies were found.

The mucous glands contained large numbers of similar bodies. They showed excessive secretion of mucus and advanced degeneration of many of the cells. The submucous and peribronchial tissues were infiltrated with mononuclear cells. No polymorphonuclear leukocytes were observed. Microscopic hemorrhages were numerous.

(*b*) Upper Lobe of Left Lung: Section of this lobe, including the consolidated area, showed part of the wall of a large bronchus. Where the epithelial surface was visible the epithelium appeared to be replaced by necrotic amorphous material in which were some degenerated nuclei. Beneath this there were dilated blood vessels lying in edematous tissue infiltrated with mononuclear

1. Brown, J. W.; Hein, G. E.; Ellman, P., and Joules, H.: Proc. Roy. Soc. Med. **36**:385, 1943.

2. Adams, J. M.: J. A. M. A. **116**:925, 1941.

cells. The mucous glands were enormously hypertrophied. The lumen contained much mucus. The lining cells showed considerable degeneration. Some of them contained masses of granules varying somewhat in size. The granules were usually sharply defined, stained red or bluish red, sometimes blue; many of them resembled the inclusion bodies found in the bronchial mucosa. Some of the few epithelial cells remaining in the bronchus presented inclusion bodies, and similar bodies were found in the necrotic areas in the lung. Some were free; some were found in large mononuclear cells.

The adjacent lung was consolidated, but the consolidation was not of the usual type. The alveoli were hard to make out. In some places they appeared to have been destroyed. Elsewhere their site was marked by a dilated capillary, but the walls of the capillary were very indistinct. Most of the area was occupied by faintly staining fibrinous material, in which were seen poorly defined cells with large nuclei derived probably from alveolar cells. Several small areas of complete necrosis were visible in this part of the lung. Some of these consisted of fibrin with a few red cells, and some, of the cells already described. Other areas showed broken-up nuclei and more nearly resembled miliary abscesses. Polymorphonuclear leukocytes, however, were hard to find. Red blood cells were fairly numerous in these areas.

Adjacent to the consolidated area the alveoli were distended, often ruptured and filled with a more or less homogeneous pink-staining material. The condition here resembled ordinary edema except that the alveolar walls were generally necrotic. The picture was like that seen in lungs in the 1918 epidemic of influenza. Collections of lymphocytes, probably derived from the normal lymphoid tissue of the lung, were seen here and there. Beyond the "edematous" area the alveoli were emphysematous and contained a few red cells and macrophages. The capillaries were intensely congested.

(*c*) Lower Lobe of Right Lung: The pleura was thickened by edema and old fibrosis. There were scattered areas of hemorrhage, and the vessels were intensely congested. The lung was extremely emphysematous and intensely congested. The emphysema often appeared acute rather than chronic. The ends of the broken alveolar walls showed curious acellular pink-staining knobs, probably consisting of inspissated fibrin. The walls of some of the alveoli were lined here and there by similar material. This may have been material similar to that found in cases of influenza, but it was present in masses rather than as hyaline membrane. Partly owing to this inspissated fibrinous material, partly to intense capillary dilatation and partly to proliferation of alveolar cells, the alveolar walls were thicker than normal. Many of the alveoli contained red cells; others were filled with edema fluid with or without mononuclear cells and blood cells. In one or two places this inflammatory edema fluid filled only part of the alveolus. It seemed to be pushed toward the alveolar wall on one side. This would explain the formation of the fibrinous clumps just described. There was little true consolidation in this part of the lung, but there were a few small areas where the alveoli were not air bearing and were filled with alveolar cells, edema fluid, blood cells and a few elongated cells resembling fibroblasts. Bronchi were difficult to distinguish; they could be located only by occasional remains of bronchial epithelium. No inclusion bodies were found in this section.

(*d*) Heart: The muscle fibers showed severe degeneration. The nuclei were often pyknotic; outlines of the cells were irregular; the cell structure was smudgy. Cross striation was generally absent. Exudate was present in the fascial planes and between muscle bundles. The exudate consisted of fibrin, which tended to stain bluish, and mononuclear cells. Red blood cells were also present, but no polymorphonuclears. The threads of fibrin were very fine, but all along them were minute sharply defined granules, sometimes pink, sometimes bluish. Bodies resembling those seen in the lung were found in the protoplasm of histiocytes and of a few muscle cells. In one place several Aschoff cells were seen around a blood vessel. All the vessels were intensely congested.

(*e*) Liver: This organ was moderately congested. The cells showed cloudy swelling and moderate fatty degeneration. There was slight round cell infiltration of the portal spaces. No inclusion bodies were seen.

(*f*) Pancreas: There was slight fatty infiltration of the pancreas. The lobules showed rather extensive areas of recent degeneration and necrosis. In some of these the walls of blood vessels were also necrotic and their contents thrombosed. Bodies resembling those in the bronchus were found in the protoplasm of some of the cells. They were surrounded by a halo and occurred singly. No secretory granules were found in cells in the areas where inclusion bodies were seen.

(*g*) Kidneys: Both kidneys were intensely congested. The cells of the tubules showed cloudy swelling, and the lumens were filled with debris. No inclusion bodies were seen. The blood vessels showed only minimal lesions.

(*h*) Spleen: The malpighian bodies were small. The pulp showed areas of congestion and hemorrhage and slight hypertrophy of littoral cells. A few of these contained inclusion bodies surrounded by a halo, apparently similar to those in the bronchus.

(*i*) Brain: Portions were fixed in alcohol, formaldehyde solution and ammonium bromide–formaldehyde solution (Hortega). The remainder were hardened in solution of formaldehyde. Paraffin blocks were made from several cortical regions, including the motor and sensory areas, the occipital lobe, the hippocampus and the cortex around the island of Reil. Serial blocks were also made on the left side through a slice comprising the insula, the claustrum, the caudate and lenticular nuclei and the lateral ventricle. The region of the right lenticular nucleus and sections through the posterior colliculus, the middle and a region near the lower end of the fourth ventricle, and the cerebellum were also examined. In addition many frozen sections were cut for a study of neuroglia and a few free hand for Nissl's original method. Stains used were hematoxylin-eosin, toluidine blue, Giemsa, Bielschowsky, Cajal's gold chloride, for neuroglia, and modifications of Hortega's stain for microglia.

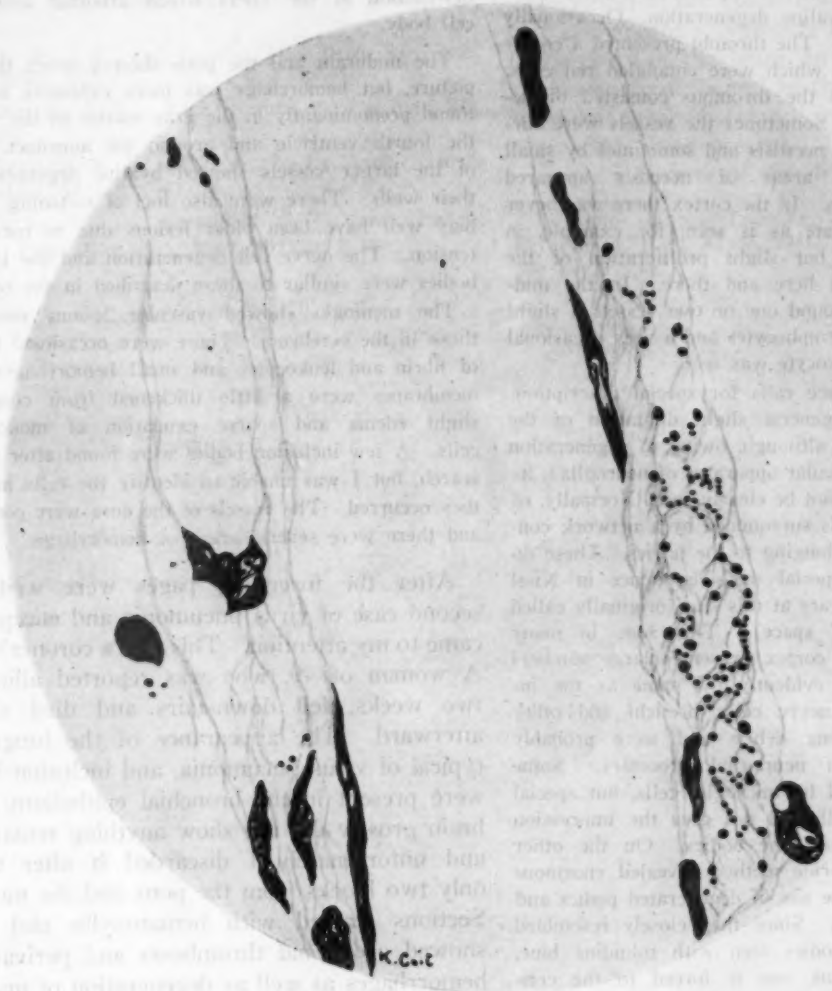
Since the same type of change was found in practically all sections, the rolandic area of the cortex will be described in detail, with only brief comment on other regions.

Sections of the cortex in the region of the fissure of Rolando showed that every nerve cell had degenerated. The Nissl granules when present were reduced to dust or clumped toward the periphery of the cells. Most of the cells showed "ring" degeneration of the protoplasm. The larger cells presented marked pigmentary degeneration; yellow pigment was often seen in the smaller cells also. Besides the pigment there were in the protoplasm and often in the nucleus of the nerve cells numbers of spherical bodies varying somewhat in size. They resembled the inclusion bodies seen in the

bronchial mucosa but were more numerous and usually smaller. Most were surrounded by a halo. Similar bodies often lay in the surrounding tissue. They appeared to have escaped from degenerated cells, but it is possible that some of them were in reality included in neuroglia cells. Sometimes a group of these bodies marked the spot where nerve cells had been. The nuclei of the nerve cells were usually pyknotic, their outline was irregular, sometimes the nuclear membrane was folded and often they lay to one side of the cell.

There was an extreme degree of neuronophagia. Also numbers of minute faintly staining particles were seen, which were probably microglia cells taking a faint toluidine stain.

Neuroglia stains revealed hypertrophy. Fibrous astrocytes with two or three nuclei were fairly common. Most of the neuroglia showed clasmatodendrosis, and in most of them the perivascular process was degenerated, but where these processes were still intact they were seen to be loaded with gliosomes. The gliosomes



Virus encephalitis. Bodies similar to those in the protoplasm of the bronchial epithelium and in nerve cells are seen in the perivascular space of a small venule.

In the more degenerated nerve cells the nuclear membrane showed incrustations. The nucleoli were generally polychromatic; sometimes the center showed a clear area. The normal clear zone around the nucleolus was filled with fine particles. Some of these were probably chromatin material, but others took a lighter reddish stain and were surrounded by a clear area or halo which was bounded by a very fine line. These structures were best seen in large cells of the motor type. In the cortex they were apt to be obscured by other particles and were therefore difficult to identify.

were large, very numerous and easily visible all the way to the cell body, where they were present in large numbers. This was true of many of the fibrous as well as of the protoplasmatic astrocytes. The nuclei of the astrocytes were increased in size, as was the cell protoplasm. In Nissl sections the nuclear membrane was sometimes encrusted. In some glia cells, bodies like those described in the nerve cells were seen close to the processes and cell body. They were much larger than gliosomes. Sometimes they seemed to be attached to the fibrils of the perivascular apparatus.

As has been said, microglia cells could be dimly made out in Nissl preparations. With silver impregnation they stained with difficulty and were for the most part minute. However moderately hypertrophied Hortega cells were found in large numbers around foci of necrosis, and some along the walls of blood vessels. Some of the sections were counterstained with sudan IV. Only two fat granule cells were seen, both near a necrotic focus in the medulla. It is probable that death intervened before the Hortega cells could attain their full development.

The blood vessels were everywhere dilated; often their walls showed hyaline degeneration. Occasionally they were thrombosed. The thrombi presented a coarse fibrinous meshwork in which were entangled red cells. In one or two places the thrombus consisted of degenerated monocytes. Sometimes the vessels were surrounded by an area of necrosis and sometimes by small hemorrhages. Some areas of necrosis appeared independently of vessels. In the cortex there was never any perivascular exudate as is seen, for example, in epidemic encephalitis, but slight proliferation of the adventitia was present here and there. In the midbrain and the pons, around one or two vessels a slight exudate consisting of lymphocytes and a very occasional polymorphonuclear leukocyte was seen.

The perivascular space calls for special description. There seemed to be general slight dilatation of the true perivascular space although, owing to degeneration of the podics [3] (perivascular apparatus of neuroglia), its boundaries often could not be clearly seen. Normally, of course, the true space is surrounded by a network consisting of fine fibrils belonging to the podics. These do not stain except by special methods, hence in Nissl preparations a gap appears at this site (originally called "false" or "shrinkage" space). This zone in many places, especially in the cortex, presented large numbers of bodies which were evidently the same as the inclusion bodies of the nerve cells, bronchi and other sites. These lay among debris and were probably originally contained in neuroglial processes. Some may have been released by microglia cells, but special stains for microglia cells did not give the impression that these cells contained the bodies. On the other hand, Cajal's gold chloride method revealed enormous numbers of bodies at the site of degenerated podics and in glia and nerve cells. Since they closely resembled in site and size the bodies seen with toluidine blue, Giemsa and other stains, one is forced to the conclusion that they were the same structures. A few bodies could be made out in the true perivascular space.

Sections (hematoxylin-eosin, Bielschowsky) through the left insula, the claustrum, the caudate and lenticular nuclei to the lateral ventricle showed vascular dilatation, fibrin and leukocytic thrombi and occasional microscopic hemorrhages. There were areas of necrosis, especially near the ventricle. Around vessels in the necrotic areas, and also near the ventricle, were large numbers of corpora amylacea. Bodies like those described in the cortex were present, but no special stains were made for them. The right lenticular nucleus was similar.

The vessels of the cerebellum were congested. There were hemorrhages in the meninges in some of the sulci and occasionally perivascular hemorrhages in the white matter. Bodies similar to those described in the cortex could be made out around the Purkinje cells and around blood vessels in some places. The Purkinje cells appeared to be degenerating. Sections stained by Bielschowsky's method showed commencing disintegration of the intracellular neurofibrils and also some destruction of the fibers which arborize around the cell body.

The midbrain and the pons showed much the same picture, but hemorrhage was more extensive and was found predominantly in the gray matter of the floor of the fourth ventricle and around the aqueduct. Some of the larger vessels showed hyaline degeneration of their walls. There were also foci of softening. These may well have been older lesions due to the hypertension. The nerve cell degeneration and the inclusion bodies were similar to those described in the cortex.

The meninges showed vascular lesions similar to those in the cerebrum. There were occasional thrombi of fibrin and leukocytes and small hemorrhages. The membranes were a little thickened from congestion, slight edema and sparse exudation of mononuclear cells. A few inclusion bodies were found after diligent search, but I was unable to identify the cells in which they occurred. The vessels of the dura were congested, and there were several areas of hemorrhage.

After the foregoing pages were written a second case of virus pneumonia and encephalitis came to my attention. This was a coroner's case. A woman of 49, who was reported ailing for two weeks, fell downstairs and died shortly afterward. The appearance of the lungs was typical of virus pneumonia, and inclusion bodies were present in the bronchial epithelium. The brain grossly did not show anything remarkable and unfortunately I discarded it after taking only two blocks from the pons and the medulla. Sections stained with hematoxylin and eosin showed occasional thromboses and perivascular hemorrhages as well as degeneration of many of the nerve cells with characteristic inclusion bodies. The bodies were also seen in the perivascular space.

COMMENT

Many of the changes observed in the brain in the case reported in detail are common to other inflammatory conditions, but the presence of inclusion bodies similar to those found in other organs seems to stamp the form of encephalitis described here as a specific entity caused by the same agent as virus pneumonia. Somewhat similar bodies were found by Da Fano

3. Ingleby, H.: J. Roy. Microscop. Soc., 1925, p. 423.

and me [4] in epidemic encephalitis in 1919. But there are certain differences in size and staining reaction and especially in location which appear to me to differentiate them. In epidemic encephalitis the bodies, for example, were on the whole smaller and were never found in the nucleus or in the perivascular space. A type of encephalitis named "inclusion encephalitis" has been described in this country by Dawson [5] and by Akelaitis and Zeldis [6] and in England by Brain, Greenfield and Russell,[7] in which intranuclear inclusion bodies were found. Similar bodies also occur in herpetic virus infection.[8] However, all these types of encephalitis differ clinically and pathologically in so many respects from that in the present case that there could be no question of confusing them.

4. Da Fano, C., and Ingleby, H.: Proc. Roy. Soc. Med. **12**:42, 1919.

5. Dawson, J. R.: Arch. Neurol. & Psychiat. **31**: 685, 1934.

6. Akelaitis, A. J., and Zeldis, L. J.: Arch. Neurol. & Psychiat. **47**:353, 1942.

7. Brain, W. R.; Greenfield, J. G., and Russell, D. S.: Proc. Roy. Soc. Med. **36**:319, 1943.

8. Smith, M. G.; Lennette, E. H., and Reames, H. R.: Am. J. Path. **17**:55, 1941.

SUMMARY

At autopsy on a patient who died of acute encephalitis complicating virus pneumonia, inclusion bodies similar to those described by Adams were found in the epithelium of the bronchi and in other organs. Numbers of them were found in all parts of the brain, in the nerve cells and the neuroglia cells and in the perivascular zone. Vascular thrombosis and acute degeneration of nerve cells were present. Perivascular exudate was not a feature of the disease.

ANOMALOUS PULMONARY VEINS

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Numerous cases of abnormal connections between the pulmonary and the systemic veins have been recorded in the literature. Such abnormal connections result in the shunting of a certain amount of oxygenated blood from the systemic circulation so that it is not available to the tissues of the body. To some extent connections between the pulmonary and the systemic veins are considered to be normal. This is evidenced by the many capillary connections between the pulmonary veins and the superior vena cava via the anterior bronchial veins, the posterior bronchial veins and the anterior mediastinal veins. To a lesser extent, the pulmonary veins are connected with the aortic venous plexus and the azygos veins via the esophageal, the pericardial and the posterior mediastinal veins. However, when a large vein drains oxygenated blood to the systemic veins or to the right atrium, the condition is anomalous. Two cases of such drainage were found in the dissecting room.

The objects of this paper are to describe these 2 cases, give a method of calculating the amount of arterial blood shunted from the general circulation and add 7 cases found in the literature to the excellent summary of anomalous pulmonary veins made recently by Brody.[1]

REPORT OF CASES

CASE 1.—Necropsy of a man 55 years old revealed the lungs and the great vessels to be normal except for a vein from the superior lobe of the right lung. It was 1 cm. long, 1 cm. in diameter and opened directly into the right side of the superior vena cava 2.5 cm. from the latter's termination in the right antrum (fig. 1). Further dissection revealed that most of the tissue of the superior lobe of the right lung was drained by this anomalous vessel. The heart was enlarged and the left ventricular wall measured 15 mm. in thickness; the walls of the other chambers were of normal thickness. There was mitral stenosis, with calcification and atheromatous plaques at the aortic valve. The tricuspid valve was normal. The circumference of the pulmonic orifice measured 8 cm., while the aortic orifice measured 9 cm.

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The specimens of anomalous pulmonary veins described in this article were dissected in the gross anatomy laboratory at Loyola Medical School. The clinical records of the patients were obtained from Cook County Hospital.

1. Brody, H.: Arch. Path. **33**: 221, 1942.

It was apparent from the clinical and postmortem observations that death was due to cardiac decompensation on the basis of rheumatic heart disease. No symptoms which might have been caused by the venous anomaly were recorded.

CASE 2.—At necropsy of a man aged 44 the lungs and great vessels were normal except for a venous communication between the superior lobe of the left lung and the left innominate vein (fig. 2). The anomalous vein was 3.5 cm. in length and 1.5 cm. in diameter. It ran ventral and to the left of the common carotid artery. It opened into the left innominate vein 4.5 cm. before the latter's junction with the right innominate vein. Further dissection revealed that almost all of the tissue of the superior lobe of the left lung was drained by the anomalous vessel (fig. 3). In the heart, mitral stenosis and hardened aortic valves were observed. The circumference of the pulmonic orifice was 9 cm., compared with a circumference of 8 cm. for the aortic orifice. The walls of the heart were of normal thickness. In this case, as in case 1, the cause of death was cardiac decompensation on the basis of rheumatic heart disease.

A METHOD OF CALCULATING THE AMOUNT OF THE SHUNTED BLOOD

In a review of the literature, no cases were found in which the actual cross-sectional area of the anomalous vessel was compared with the total cross-sectional area of the veins normally draining the lung tissue. Approximations of the amount of lung tissue abnormally drained have been made by several authors, but there are no records of measurements having been taken.

In case 1 the total cross-sectional area of all pulmonary veins was 409 sq. mm. while that of the anomalous vessel was 113 sq. mm. This indicates that 26.1 per cent of the blood oxygenated in the lungs was shunted to the right side of the heart via the superior vena cava. Thus it is apparent that approximately one fourth of the lung tissue was nonfunctional so far as the needs of the tissues of the body for oxygen were concerned.

In case 2 the total cross-sectional area was 617 sq. mm. and that of the anomalous pulmonary vein was 124 sq. mm. Thus 20.1 per cent of oxygenated blood was not available to the tissues of the body.

COMMENT

Proper recognition has been given the excellent résumé of the literature made recently by Brody.[1] His impression that anomalous drain-

age of less than half of the pulmonary blood is compatible with life at least into adulthood agrees with the impression received by us in a study of the literature.

To his collection of 106 cases of anomalous drainage of all or part of the lungs should be added the 2 cases just reported and also the following 7 cases:

1. Heller[2]: The superior pulmonary vein drained into the superior vena cava.

2. Ingalls[3]: All pulmonary veins joined to form a common trunk which drained into the superior vena cava. Associated anomalies were interventricular and interatrial defects, pulmonary arteries branching from the aorta and dextrocardia. These anomalies were seen in an 8 week old girl, whose death was ascribed to bronchopneumonia.

3. Konaschko[4]: Two right pulmonary veins emptied into the superior vena cava.

4. Töply[5]: All pulmonary veins on the left drained into the superior vena cava except two very small ones which emptied into the left ventricle. Other postmortem observations included hypertrophy of the left atrium and ventricle and mitral insufficiency. No mention is made of a septal defect. This anomaly occurred in a 20 year old man, whose death was ascribed to lobar pneumonia and cardiac decompensation.

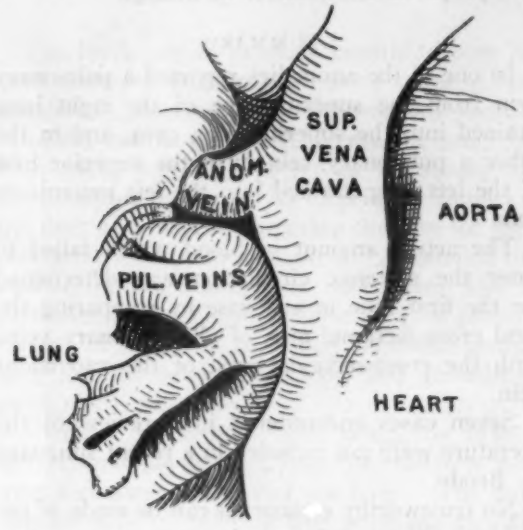

Fig. 1.—Diagram illustrating the anomalous vein in case 1.

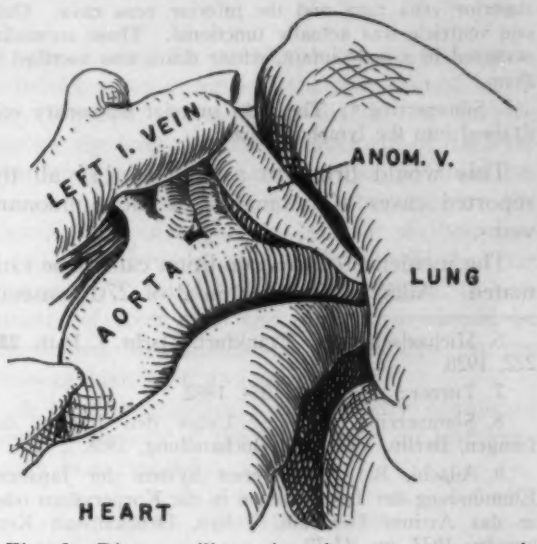

Fig. 2.—Diagram illustrating the venous communication between the superior lobe of the left lung and the left innominate vein in case 2.

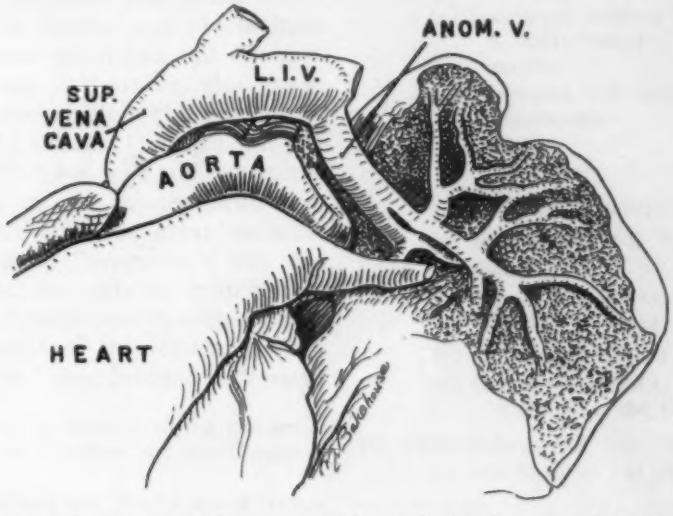

Fig. 3.—Diagram showing that in case 2 the superior lobe of the left lung was drained chiefly by the anomalous vein. *L. I. V.* indicates the left innominate vein.

2. Heller: Verhandl. d. deutsch. path. Gesellsch. **12**:248, 1908.

3. Ingalls, N. W.: Anat. Rec. **53**:269, 1932.

4. Konaschko, P. I.: Ztschr. f. Anat. u. Entwcklngsgesch. **89**:672, 1929.

5. Töply, R.: Prag. med. Wchnschr. **7**:233, 1882.

5. Michaelsohn [6]: All pulmonary veins drained into a persistent left superior vena cava which emptied into the left atrium. Associated anomalies were a bilocular heart and situs inversus. These anomalies occurred in a 21 year old man, whose death was ascribed to them.

6. Turner [7]: All pulmonary veins drained into a persistent left superior vena cava which emptied into single atrium. This chamber also received the right superior vena cava and the inferior vena cava. Only one ventricle was actually functional. These anomalies occurred in a male infant, whose death was ascribed to them.

7. Sömmerring [8]: The right superior pulmonary vein drained into the lymphatic duct.

This would bring to a total of 115 all the reported cases of anomalies of the pulmonary veins.

The incidence of this condition cannot be estimated. Adachi [9] found it in 2 of 270 consecutively examined cadavers, an incidence of 0.7 per cent. Of 280 cadavers examined by us, only 2 (cases 1 and 2 in this report) showed the anomaly, also an incidence of 0.7 per cent. Neither study, however, was extensive and neither included examination of infants, children or a proportionate number of women.

6. Michaelsohn, A.: Frankfurt. Ztschr. f. Path. **23**: 222, 1920.

7. Turner: Lancet **2**:1034, 1882.

8. Sömmerring, S. T.: Ueber den Bahnen der Lungen, Berlin, Vossische Buchandlung, 1808.

9. Adachi, B.: Das Venen System der Japaner: Einmündung der Lungenvenen in die Korpervenen oder in das Atrium Dextrum, Tokyo, Druckanstalt Kenkyusha, 1933, pp. 33-39.

SUMMARY

In one of the anomalies reported a pulmonary vein from the superior lobe of the right lung drained into the superior vena cava, and in the other a pulmonary vein from the superior lobe of the left lung drained into the left innominate vein.

The actual amount of blood which failed to enter the systemic circulation was determined for the first time in any case by comparing the total cross-sectional area of all pulmonary veins with the cross-sectional area of the anomalous vein.

Seven cases encountered in a review of the literature were not included in a recent summary by Brody.

No trustworthy estimation can be made of the incidence of anomalous pulmonary veins.

PRIMARY NEOPLASMS OF THE LIVER

WESLEY N. WARVI, M.D.*

CINCINNATI

The literature on primary hepatic tumors has been reviewed and an analysis of cases of such neoplasms observed at the Cincinnati General Hospital over a period of twenty-six years has been made with the object of facilitating the classification and diagnosis of primary tumors of the liver and thereby preparing the way for early corrective therapeutic measures. A detailed discussion of the therapeutic procedures will be published later. Hepatocellular types of primary tumors of the liver are given emphasis because they are the least understood and accordingly most often mistreated.

Virchow is credited with being the first to differentiate the primary from the relatively common secondary tumors of the liver. The more delicate task of defining true adenoma and distinguishing it from simple compensatory hyperplastic nodules was undertaken by Simmonds.[1] From the point of view of gross examination, Hanot and Gilbert[2] proposed that primary carcinoma of the liver be divided into (1) nodular carcinoma, (2) massive carcinoma and (3) carcinoma with cirrhosis. However, this gross classification was incompatible with their microscopic division of (1) alveolar (biliary) and (2) trabecular (liver cell) types. In addition to the confusing classification, the nomenclature has added to the difficulties by the great variation in the use of the word "hepatoma"; this has been applied to structures varying from benign nodules to highly malignant cancers with or without associated cirrhosis of the remaining liver tissue.[3] Some authors[4] have justified such usage by questioning the neoplastic nature of hepatocellular tumors in view of the retention of secretory function, the apparent inability to survive outside the liver and the involvement of the liver from multicentric foci rather than by invasion. Brines[5] included bile duct tumors under the classification of hepatoma because of their histogenic relationship. It is believed that the term "hepatoma" should be sharply restricted to true tumors of liver cells and that these should be subdivided into malignant and benign on the basis of histologic and other data.

A brief classification of clinically significant primary tumors of the liver based on histologic study is presented:

TRUE PRIMARY TUMORS

I. Hepatoma
   1. Liver cell (or hepatocellular) adenoma ("trabecular adenoma")
   2. Liver cell (or hepatocellular) carcinoma
      (*a*) Carcinoma without cirrhosis, occurring as a single "massive" growth or as multiple nodules
      (*b*) Carcinoma with cirrhosis, almost invariably multinodular

II. Cholangioma
   1. Adenoma
      (*a*) Solid (or "tubular") type
      (*b*) Cystadenoma (or "vesicular") type
   2. Carcinoma
      (*a*) Adenocarcinoma, varying from alveolar to medullary forms
      (*b*) Cystadenocarcinoma
      (*c*) Carcinoma simplex, usually associated with cirrhosis of the liver

III. Cholangiohepatoma (or "mixed" tumor), exhibiting both bile duct and liver parenchyma cells

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* At the time of writing Dr. Warvi was a trainee in the diagnosis and treatment of cancer of the National Cancer Institute.

1. Simmonds, M.: Deutsches Arch. f. klin. Med. **34**:388, 1884.

2. Hanot, V., and Gilbert, A.: Etudes sur les maladies du foie, Paris, Asselin & Houseau, 1888, p. 334.

3. (*a*) Cabot Case 23051, New England J. Med. **216**:209, 1937. (*b*) Cathala, J.: Paris méd. **49**:508, 1923. (*c*) Eggel, H.: Beitr. z. path. Anat. u. z. allg. Path. **30**:506, 1901. (*d*) Ewing, J.: Neoplastic Diseases, ed. 4, Philadelphia, W. B. Saunders Company, 1940. (*e*) Fischer, H.: Ann. Surg. **35**:467, 1927. (*f*) Fried, P. M.: Am. J. M. Sc. **168**:241, 1924. (*g*) Glennon, W. P., and Byona, R. V.: J. Missouri M A. **30**:142, 1933. (*h*) Rolleston, H. D., and McNee, J. W.: Diseases of Liver, Gallbladder and Bile Ducts, ed. 3, New York, The Macmillan Company, 1929, p. 478. (*i*) Shaw, A. F. B.: J. Path. & Bact. **26**: 475, 1923. (*j*) Smith, L. W.: Arch. Path. **1**:365, 1926. (*k*) Tull, J. C.: J. Path. & Bact. **35**:557, 1932. (*l*) Witwicky, R.: Ztschr. f. klin. Med. **36**:475, 1899. (*m*) Yamagiwa, K.: Virchows Arch. f. path. Anat. **206**:437, 1911.

4. Géraudel, E.: J. de l'anat. et physiol. **43**:410, 1907; cited by Ewing,[3d] p. 748.

5. Brines, O. A.: Am. J. Clin. Path. **3**:221, 1933.

IV. Tumors primary in the liver but not containing specific hepatic elements

1. Tumors of vascular origin—hemangioma, lymphangioma, endothelioma
2. "Adrenal rest" tumor
3. Sarcoma
4. Teratoid tumor
5. (?) Primary malignant melanoma of the liver

Certain abnormal types of hyperplasia have been incorrectly diagnosed as neoplasms. The physiologic regeneration of traumatized liver cells is usually orderly and easily recognized, but irregular, diffuse or nodular types of hyperplasia have been confused with new growths. Diffuse hyperplasia associated with diabetes or von Gierke's disease might similarly be misinterpreted as neoplastic rather than metabolic because of the nuclear distortion caused by imbibition of glycogen. The nodular hyperplasia of cirrhosis, erroneously called "multiple adenoma," fails to fulfil the criteria of true adenoma: The involved areas do not possess true capsules but are enveloped in condensations of the fibrous tissue which permeates the cirrhotic liver. The zones of regeneration represent attempted restoration of liver parenchyma rather than autogenous foci. These cell groups are capable of secreting bile, which passes into the imperfect canaliculi connected to the normal duct system. They may become neoplastic, for hepatocellular cancers statistically occur far more often in cirrhotic than in normal livers. Exclusion of such hyperplastic nodules greatly reduces the number of tumors reported as adenoma.

True liver cell adenoma is an encapsulated tumor probably of congenital origin, usually single, with complete anatomic and physiologic isolation from normal liver tissue. It is among the rarest tumors of specific hepatic elements. Its incidence will be discussed later. The clinical limits of this class of tumors are controversial, for some authors [6] include only the benign neoplasms which they believe never become cancers, while others [7] describe all hepatocellular tumors, including adenoma, as actually cancers. Cathala [3b] straddled the issue by classifying both benign and cancerous tumors of liver cells as adenoma. Some authors (Greenfield [7d]; Toupet [7n]; Varadi [7p]) limit the classification of adenoma to tumors that histologically appear benign but show invasion of blood vessels and dissemination by the blood stream. Varadi [7p] diagnosed a tumor as liver cell adenoma on biopsy of a metastatic lesion in bone. These authors reason that the structural uniformity of tumors arising in a vascular organ such as the liver is analogous to that of tumors diagnosed as thyroid adenoma which exhibit invasion of blood vessels and distant deposits. It seems more reasonable to classify hepatocellular tumors showing invasion of blood vessels and metastasis as carcinoma, regardless of their regularity and benign histologic appearance. The most costly error in classification is the grouping of adenoma as carcinoma, for the latter generally is regarded as incurable and treated purely symptomatically; thus the patient with adenoma is deprived of any chance of recovery. This misunderstanding is clearly illustrated in Peugniez' [7l] use of the contradictory term "primary massive encapsulated carcinoma." The mistaken identification of carcinoma as adenoma appears less serious in view of the mild surgical risk of exploration, which is justified in view of the rapidly fatal prognosis for untreated carcinoma. Authors [8] who have made careful studies of this subject and have reported cases of their own are of the opinion, in which I concur, that liver cell tumors to be called adenoma must be benign in all respects, but the potentiality of cancerous transformation is always present. The relationship of the liver cell tumor to hamartoma is at present poorly understood. The latter has been defined as a congenital tumor-like malformation characterized by a relative quantitative disproportion of the various tissue components normally comprising a given organ. I have not placed it as a distinct or separate type among the tumors of the liver, as in most cases it appears to be composed solely or predominantly of liver cords and very often is not distinguishable on histologic grounds from adenoma. Thus this indolent nodule, 5 examples of which I have observed, is included among the tumors diagnosed as adenoma; in all cases it shows liver

6. (*a*) Dévé, F.: Normandie méd. **29**:157, 1913. (*b*) Lucène, P.: Rev. de gynéc. et de chir. abd. **19**: 555, 1912. (*c*) McRae, F. W.: Am. J. Surg. **28**:575, 1935. (*d*) Mongé, J.: Bordeaux chir. **10**:42, 1939.

7. (*a*) Aubertin, C.: Bull. et mém. Soc. méd. d. hôp. de Paris **46**:818, 1930. (*b*) Birch-Hirschfeld, F. V., in Gerhardt, C.: Handbuch der Kinderkrankheiten, Tübingen, H. Laupp, 1880, vol. 4, pt. 2, p. 825. (*c*) Goffin, R.: J. de chir. et ann. Soc. belge de chir. **35-37**:157, 1938. (*d*) Greenfield, J.: Arch. d. Heilk. **5**:261, 1864. (*e*) Griffith, J. P. C.: Am. J. M. Sc. **155**:79, 1918. (*f*) Hicks, E. S.: Canad. M. A. J. **20**:169, 1929. (*g*) Karsner, H. T.: Arch. Int. Med. **8**:238, 1911. (*h*) Mahomed, F. A.: Tr. Path. Soc. London **28**:144, 1876-1877. (*i*) Nadal, P.: Bull. et mém. Soc. anat. de Paris **88**:355, 1913. (*j*) Pepere, A.: Arch. per le sc. med. **26**:117, 1902. (*k*) Perls, M.: Virchows Arch. f. path. Anat. **56**:435, 1872. (*l*) Peugniez, P.: Bull. Soc. anat. de Paris **77**:456, 1902. (*m*) Rénon, L.; Géraudel, E., and Monier-Vinard, R.: Arch. de méd. expér. et d'anat. path. **22**:311, 1910. (*n*) Toupet: J. de méd. de Paris **41**:879, 1922. (*o*) Tuholske, H.: Am. J. Surg. **26**:52, 1912. (*p*) Varadi, S.: Sang **11**:872, 1937. (*q*) Wallace, R. H.: Arch. Surg. **43**: 14, 1941.

8. Ewing.[3d] Perls.[7k] Greenfield.[7d] Tuholske.[7o]

cells in cordlike arrangement without bile duct formation and is demarcated from the rest of the liver by an irregular, usually incomplete fibrous membrane varying from an almost imperceptible amount of tissue to a dense connective tissue capsule in the cases in which it attains a large size. Histogenetically, physiologically and anatomically hamartoma fulfils most or all of the criteria for the diagnosis of hepatocellular adenoma, and until finer diagnostic methods are available, it should be classified as such; both frequently appear to be congenital, both fail to have connections to the duct system of the liver or to take part in its functions, and both are composed of adult liver cells attempting to reproduce the corded architecture of the gland but are lacking in portal triads or bile canaliculi. Warthin [9] claimed that hamartoma differs from adenoma in its quiescence, calling it the product of inhibitory rather than proliferative aberration. However, when adenoma of liver cells first appears in adult life it must be assumed to have remained quiescent all during childhood and youth and to have proliferated later to the point of clinical manifestations, as reported by several authors. [10] Rolleston and McNee [3h] expressed the belief that liver cell adenoma is always hamartomatous. This is also said to be the opinion of Dyrenforth [11] in a publication on the histogenesis of hepatocellular tumors, but I have been unable to confirm the existence of this publication.

I have grouped tumors diagnosed as liver cell carcinoma (or malignant hepatoma) in two divisions dependent on the presence or the absence of cirrhosis. It is the latter factor that appears significant in the clinical course and prognosis of the disease process. In the presence of cirrhosis a variable history of chronic or low grade hepatitis may be elicited, often on an alcoholic or a dietary basis. Anatomically, hyperplastic liver cell nodules gradually fade into carcinomatous tissue in numerous areas throughout the diffusely nodular liver, offering no possibility of successful operative intervention. Carcinoma in the form of a single massive growth offers a better prognosis from the therapeutic angle in spite of its shorter course, as the rest of the liver usually is physiologically normal and frequently permits resection of the tumor mass. In many cases of this type carcinoma appears to arise from adenoma of the liver cells and the lesion exhibits all degrees of change from benign adenoma with slight cancerous transformation to highly anaplastic carcinoma. Highly undifferentiated carcinoma in the form of a single massive growth, however, is rarely found in the absence of other intrahepatic carcinomatous tumors. Many authors [12] divide hepatic carcinoma into three types as proposed by Nicholls, Hektoen and Riesman [12e] in 1901: carcinoma in the form of a single massive growth, the most amenable to treatment: the multiple nodular form without cirrhosis, and the multiple nodular form with cirrhosis, the last being most frequent and having the poorest prognosis.

Cholangioma is a tumor of biliary duct origin. Noncancerous types comprise the rare solid or "tubular" types and the relatively common type diagnosed as cystadenoma or the "vesicular" type. Cystadenoma of the liver is histologically similar to cystadenoma of any other gland, such as the ovary. It must be differentiated from simple retention cyst and from generalized cystic disease, affecting also the kidneys, the pancreas, the spleen and other organs. Cancerous cystadenoma may merge into medullary types of carcinoma with cyst formation. Cancerous growth in a cirrhotic liver may arise either from bile ducts or from liver cells and usually results in carcinoma simplex with a rapid course leading to death in one to six weeks.

Cholangiohepatoma (often confusingly called "mixed tumor" [3d]) is simultaneous neoplastic proliferation of bile duct cells and liver parenchyma cells with predominance of the bile duct character, for the latter element appears to grow and spread more rapidly. Distinct tumors of this type are uncommon, for one of the elements usually comprises a sufficient portion of the growth to obviate this diagnosis.

The classification of primary tumors of the liver other than those arising from specific hepatic elements depends on their origin from associated mesenchymal structures. Tumors of vascular origin, particularly those designated as hemangioma, make up the greater part; they vary in size from 0.5 to 15 cm. or more in diameter and

9. Warthin, A. S., in Buck, A. H.: Reference Handbook of Medical Sciences, New York, William Wood & Company, 1923, vol. 4, p. 855.

10. Albrecht, E.: Verhandl. d. deutsch. path. Gesellsch. **7**:153, 1904. Christiani: J. d. l'anat. et physiol. **27**:271, 1891. Sanford, D. A.: J. Obst. & Gynaec. Brit. Emp. **48**:246, 1941. Schrager, V. L.: Ann. Surg. **105**:33, 1937. Wagner, E.: Arch. d. Heilk. **1**:251, 1861. Warthin.[9]

11. Dyrenforth, L. C., cited by Smith.[27]

12. (*a*) Abel, A. L.: Brit. J. Surg. **21**:684, 1934; Tr. M. Soc. London **60**:136, 1937. (*b*) Briggs, J. F.: Minnesota Med. **16**:230, 1933. (*c*) Glynn, E.: Brit. M. J. **2**:1192, 1911. (*d*) Muir, R.: J. Path. & Bact. **12**: 287, 1908. (*e*) Nicholls, A.; Hekteon, L., and Riesman, D.: An American Textbook of Pathology, Philadelphia, W. B. Saunders Company, 1901, vol. 2, p. 815. (*f*) Rosenbusch, H.: Virchows Arch. f. path. Anat. **261**: 326, 1926. (*g*) Sabourin, C.: Rev. de méd. **4**:321, 1884. (*h*) Schiwoff, M.: Zur Histogenese des primären Lebercarcinoms, Zurich, Leemann & Co., 1918. (*i*) Stewart, M. J.: Lancet **2**:565, 617 and 669, 1931.

range from spongy to almost solid consistency. The latter form comprises transitions from the more anaplastic angioendothelioma to the highly malignant endothelioma. The more cellular anaplastic tumors described as mesenchymal in origin are not accepted universally because of the difficulty in differentiating them from highly malignant carcinoma. Tumors acceptable as angioendothelioma, endothelioma or sarcoma are eliminated as carcinoma by other authorities.[13] Uncertainty is reflected in the reports [14] of tumors having both carcinomatous and sarcomatous elements intermingled or in separate portions. Both may show pleomorphism, giant cells and marked vascularity, but if granularity of cytoplasm is noted, the tumor should be classed as carcinoma of liver cells. An interesting stromagenous tumor probably arising from the mesodermal primordium was reported by Foot.[15] In considering "adrenal rest" tumors of the liver, such as that reported by Rolleston and McNee,[3h] one must remember the histologic similarity caused by glycogen and lipoid vacuolation of cells composing primary parenchymal tumors of the liver. Both benign and cancerous tumors having their origin in adrenal rests have been reported.[16] I have found no tumor of this type in the liver. Teratomatous mixed tumors have been reported,[17] but these forms occur less frequently in the liver than in other parts of the body. Castle [18] reported a case of adenocarcinoma arising within teratoma of the liver. The most questionable type of primary tumor of the liver reported is that diagnosed as malignant melanoma. Three authors, Koch,[19] Brandt [20] and Ascoli [13a] claimed to have studied histologically examples of primary melanoma of the liver which could withstand criticism. Brandt [20] expressed the belief that the long dendrite-like processes of the tumor cells in his case showed a relationship to the Kupffer cells. I have not observed any indisputable case of melanoma primary in the liver. Henke and Lubarsch [21] stressed easily overlooked or forgotten small lesions of the eyes, the skin or the brain that have been eradicated surgically or by irradiation as likely sources of the so-called primary melanoma of the liver.

13. (*a*) Ascoli, V.: Policlinico (sez. chir.) **37**:309, 1930. (*b*) Jaffé, R. H.: Arch. Int. Med. **33**:330, 1924. Ewing.[3d] Brines.[5]

14. Terplan: Centralbl. f. allg. Path. u. path. Anat. **31**:453, 1920.

15. Foot, N. C.: Am. J. Path. **3**:653, 1927.

16. (*a*) Flemming, A. L.: Brit. M. J. **2**:1475, 1911. (*b*) Nitch, C. A. R.: Proc. Roy. Soc. Med. (Surg. Sect.) **16**:64, 1923. (*c*) Ramsey, T. L.: Ann. Surg. **90**:41, 1929.

17. Hippel, B.: Virchows Arch. f. path. Anat. **201**: 326, 1910. Philipp, P. W.: Jahrb. f. Kinderh. **68**:353, 1908. Sheehan, H. L.: J. Path. & Bact. **33**:251, 1930. Skifosovsk, N. V.: Rev. de chir. **16**:984, 1896. Yamagiwa.[3m] McRae.[6c]

18. Castle, O. L.: Surg., Gynec. & Obst. **18**:477, 1914.

19. Koch, R.: Virchows Arch. f. path. Anat. **201**: 326, 1916.

20. Brandt, M.: Ztschr. f. Krebsforsch. **31**:254, 1930.

INCIDENCE OF PRIMARY TUMORS OF THE LIVER

Knowledge of the incidence of primary tumors is of value in considering the possible occurrence of such a tumor in a case to be diagnosed. An enlargement of the liver usually represents physiologic hyperplasia and excessive regeneration rather than a true tumor. Jaffé [13b] attributed the rarity of the latter to the fact that the regenerative capacity of the liver is greater than that of any other part of the body.

Adenoma is not as rare as commonly believed. In the majority of cases it arises from bile ducts and is cystic in type. Several authors [22] have collected a hundred or more cases of cystadenoma in recent years. Ackman and Rhea [23] found cystadenoma in 0.18 per cent of all autopsies. The less frequent "solid" adenoma is of hepatocellular origin in most instances. A total of 168 cases have been reported in the literature as histologically proved, but 79 of them are cited in reviews [24] that cannot be checked for possible reduplication of the figures. Of these, only 62 have detailed microscopic descriptions compatible with true adenoma; in 38 of the 62 cases the tumor was resected surgically. In an additional 24 cases microscopic studies were made, but as these showed some evidence of cancer the cases are not acceptable. I have observed 5 cases at this institution, bringing the total to 67 microscopically proved cases of adenoma of liver cells.

Carcinoma in the liver must be considered secondary until proved primary, for cases of the former predominate by a large majority. Virchow is credited with the dictum "such organs as are frequent sites of metastatic tumor are rarely the site of original neoplasm," emphasizing the rarity of primary tumors of the liver. The ratio of cases of primary to cases of secondary carcinoma varies in many reports as follows:

21. Henke, F., and Lubarsch, O.: Handbuch der speziellen pathologischen Anatomie und Histologie, Berlin, Julius Springer, 1928, vol. 5, p. 1.

22. (*a*) Beattie, D. A., and Robertson, H. D.: Lancet **2**:674, 1932. (*b*) Montgomery, A. H.: Arch. Surg. **41**:422, 1940. (*c*) Wikle, H. T., and Charache, H.: Am. J. Surg. **31**:345, 1936.

23. Ackman, F. D., and Rhea, L. J.: Brit. J. Surg. **18**:648, 1931.

24. Caminiti, R.: Arch. f. klin. Chir. **69**:630, 1903. Gordinier, H. C., and Sawyer, H. P.: Am. J. M. Sc. **145**:258, 1913.

1 to 64.5 (Orth [25]); 1 to 40 (Hansemann [26]); 1 to 25 (K. J. Smith [27]); 1 to 20 (Lichtman [28]). The percentage of cancers of the liver that are primary is stated as 17 by L. W. Smith,[21] 8.4 by Brines,[5] 1.5 to 3.0 by Jaffé [13b] and 1.2 by Colwell.[29] The incidence of primary tumors of the liver at all autopsies was found to be 0.5 per cent by Charache,[30] 0.25 per cent by Gustafson [31] and 0.12 per cent by Allbutt and Rolleston.[32] Charache [30] stated that the total number of reported cases in 1939 was 1,125, but there was some reduplication; elimination of this reduced the total to 1,109.[33] Since that time 66 cases have been published, and I am reporting 25 cases to total 1,200 histologically studied cases up to the present time.

The majority of mesenchymal tumors primary in the liver are of no clinical significance. Peck [34] found that only 21 cases of hemangioma which called for operative removal had been reported up to 1921. Gasparian [35] found 4 cases of lymphangioma of the liver prior to 1928. Cases both of adenoma and of carcinoma with origin in an adrenal rest have been reported [36] but almost all were encountered accidentally; Schmorl,[37] for instance, stated that they were met with in 1.7 per cent of routine autopsies. Forty-eight cases of primary sarcoma were reported by Jaffé[13b] in a review in 1924, but others would lower [4] or raise [33x] this number. Four recorded cases of primary melanoma of the liver have not been disproved. One of these was observed by Koch,[19] 2 by Brandt [20] and 1 by Ascoli.[13a] Hence primary mesenchymal tumors except the inconsequential ones classified as hemangioma are uncommon and of little clinical importance.

ETIOLOGY

Etiologic factors can be grouped into general and specific, the former affecting the patient and the latter directly concerning the hepatic growth. General factors include heredity, age, sex, race and climate. Abel [12a] reported a hereditary tendency toward primary tumor of the liver. The age at which adenoma usually becomes manifest is that of childhood, with multiple cystadenoma of the liver occurring in the earliest years, single cystadenoma more in later childhood and liver cell adenoma in adolescence and early adult life. They are all presumably congenital, but cystadenoma is noted earlier because of its more rapid growth and accumulation of fluid. Primary carcinoma of the liver occurs at any age from that of the fetus [38] to the age of 83 years [39] but is

25. Orth, M.: Ueber primäres Lebersarkom, Strassburg, J. H. E. Heitz, 1885.

26. Hansemann, D.: Berl. klin. Wchnschr. **27**:353, 1890.

27. Smith, K. J.: J. Lab. & Clin. Med. **18**:915, 1933.

28. Lichtman, S. S.: Diseases of the Liver, Gallbladder and Bile Ducts, Philadelphia, Lea & Febiger, 1942, p. 572.

29. Colwell, H.: Middlesex Hosp. **5**:123, 1905.

30. Charache, H.: Am. J. Surg. **43**:96, 1939.

31. Gustafson, E. G.: Ann. Int. Med. **11**:889, 1938.

32. Allbutt, T. C., and Rolleston, H. D.: System of Medicine, London, Macmillan & Co., 1908, vol. 4, p. 215.

33. (*a*) Armand-Delille, P.; Fèvre, M., Lestocquoy, C.: Bull. Soc. de pédiat. de Paris **33**:480, 1935. (*b*) Barry, M. W., and Russum, B. C.: Nebraska M. J. **16**: 312, 1931. (*c*) von Bardeleben: Verhandl. d. deutsch. Gesellsch. f. Chir. **22**:10, 1893. (*d*) Bastianelli, P.: Arch. ital. di chir. **34**:207, 1933. (*e*) Bauer, J. L.: Long Island M. J. **10**:341, 1916. (*f*) Baumgartner, A., and Fiessinger, N.: Bull. et mém. Soc. de chirurgiens de Paris **61**:772, 1935. (*g*) Beer, E.: Ann. Surg. **89**: 780, 1929. (*h*) Bertini, G.: Boll. e mem. Soc. piemontese di chir. **8**:183, 1938. (*i*) Bucalossi, P.: Arch. ital. di chir. **31**:441, 1932. (*j*) Cagnetto, G.: Arch. per le sc. med. **34**:495, 1910. (*k*) Chalier and Martin: Bull. Soc. nat. de chir. **55**:1385, 1929. (*l*) Chiray, Brocq, Albot and Lanthier: Mem. Acad. de chir. **64**: 911, 1938. (*m*) Donzelli, L.: Cesalpino **9**:7, 1913. (*n*) Engelhardt, A.: Deutsches Arch. f. klin. Med. **60**:607, 1898. (*o*) Escher, cited by Anschütz.[70b] (*p*) Farani, A.: Arch. brasil. de med. **14**:361, 1924. (*q*) Fischer, E.: Deutsche Ztschr. f. Chir. **210**:404, 1928. (*r*) Geremia, A.: Gior. veneto di sc. med. **9**:168, 1935. (*s*) Johansson, S.: Nord. med. Ark. **50**:10, 1917. (*t*) Kolsch, L. F., and Kiener, P. L.: Arch. de physiol. **3**:622, 1876. (*u*) Kidd, F.: Proc. Roy. Soc. Med. (Sect. Surg.) **16**:61, 1922-1923. (*v*) Lilienthal, H., cited by Tinker, M. B.: Ann. Surg. **102**:728, 1935. (*w*) Lius, A.: Gazz. d. clin. **23**:225, 1886; Centralbl. f. Chir. **14**:99, 1887. (*x*) Marino: Bol. y trab. de la Soc. de cir. de Buenos Aires **8**:381, 1924. (*y*) Mason, G. A.: Univ. Durham Coll. M. Gaz. **22**:38, 1921-1922. (*z*) Nicotra, A. S.: Morgagni (pt. 1) **55**:54, 1913. (*a'*) Oliver, J. C.: Tr. South. S. & Gynec. A. **23**:38, 1911. (*b'*) Pallarés Iranzo, V., and Bartual Vincéns, L.: Crón. méd., Valencia **40**:345, 1936. (*c'*) Pettinani, V.: Bollettino **13**:95, 1931. (*d'*) Ribadeau-Dumas, L., and DeLaulerie: Bull. et mém. Soc. méd. d. hôp. de Paris **44**:1101, 1920. (*e'*) Rossi, F.: Tumori **10**:171, 1923. (*f'*) Schmidt, F.: Verhandl. d. deutsch. Gesellsch. f. Chir. **22**:9, 1893. (*g'*) Shattock, S. G.: Proc. Roy. Soc. Med. (Path. Sect.) **3**:153, 1909-1910. (*h'*) Shaw, A. F. B.: J. Path. & Bact. **26**:475, 1923. (*i'*) Siegmund, A.: Virchows Arch. f. path. Anat. **115**:155, 1899; (*j'*) St. Thomas Hosp. Rep. **33**:83, 1904, (*k'*) Szymonowicz, J.: Polski przegl. chir. **11**:31, 1932. (*l'*) Tavares, A.: Arq. de pat. **6**:520, 1934. (*m'*) Tiffany, L. M.: Maryland M. J. **23**:531, 1890. (*n'*) Tomita, I., and Kinoshita, Y.: Ztschr. d. japan. chir. Gesellsch. **35**:46, 1934. (*o'*) Turner, P.: Proc. Roy Soc. Med. (Surg. Sect.) **16**:60, 1922-1923. (*p'*) Wright, G.: ibid. **16**:56, 1922-1923.

34. Peck, C. H.: Tr. South. S. A. **33**:289, 1920-1921.

35. Gasparian, G. I.: Arch. f. klin. Chir. **153**:435, 1928.

36. Rolleston and McNee.[3h] Flemming.[16a] Nitch.[16b] Ramsey.[16c] Orth.[25]

37. Schmorl, G.: Beitr. z. path. Anat. u. z. allg. Path. **9**:524, 1891.

38. Dansie, C. B.: Lancet **2**:228, 1922.

39. Müller, W.: Zentralbl. f. allg. Path. u. path. Anat. **61**:161, 1932.

most frequent from 50 to 60 years of age.[3m] Of 77 tumors of this type tabulated by Steiner [40] as occurring in childhood 53.2 per cent occurred in persons under 2 years of age. Males are more predisposed to primary liver cell carcinoma (this apparently bears some relation to the increased incidence of cirrhosis in males) while females have a greater frequency of bile duct carcinoma, presumed to follow proliferating cholangitis secondary to intraductal inflammation. Near and Far Eastern races have a greater incidence of liver cell carcinoma than those of Europe or the Americas. This is particularly striking in Chinese and in African natives; the latter appear to have proportionately fewer tumors elsewhere in the body. This increased racial incidence at least in Chinese appears to be connected with the frequent occurrence of cirrhosis secondary to schistosomiasis.[41]

Specific factors considered etiologically significant by investigators of primary tumors of the liver are: rests of liver cells or accessory duct structures; long-standing chemical, bacterial, mechanical or toxic irritation affecting the liver cells; progressive cirrhotic changes; avitaminosis or disturbed nutrition altering the metabolism and growth of liver cells. Physiologically isolated liver cells present since birth or acquired by stricture of the tissue dependent on dress ("corset liver") proliferate to form a clinically manifest tumor.[42] Cirrhosis, however, has occupied the center of interest in the etiology of tumors of the liver. Authors have varied in their appraisal of its significance; some have expressed the belief that it is highly important and have reported its presence in 70 to 100 per cent of patients with liver cell carcinoma and in 50 per cent of those with bile duct carcinoma [43]; others [27] have expressed the belief that it is less important and have found it associated in only 39 per cent of patients. My associates and I have found cirrhosis associated in 46 per cent of our cases of primary liver cell carcinoma. The duration of the cirrhosis prior to the evidence of the presence of a neoplasm is exceedingly variable, but in general the process of transformation appears to take several years. In a case studied by me the peritoneal cavity was tapped seven times in 1918 for recurring ascitic fluid secondary to portal cirrhosis of the liver. Following this treatment the patient remained clinically well and symptom free until his illness in 1940, twenty-two years later, which resulted in his death. At autopsy there was advanced cirrhosis of the liver with multiple nodules of primary hepatocellular carcinoma scattered throughout the tissue. Cirrhosis associated with pigmentation is reported to undergo cancerous regeneration more often than cirrhosis secondary to stasis.[44] Carli [45] outlined the possible roles played by cirrhosis as follows: that in which it precedes carcinoma and is etiologic; that in which it occurs concomitant with carcinoma as a result of the same irritation that causes the cancerous change; that in which it occurs after and results from the tumor growth. Experimentally avitaminosis B has been shown to predispose to carcinoma in the presence of excessive dietary fats or the administration of carcinogenic agents,[46] and lack of vitamin A was found to leave the surviving liver cells in a state of anaplasia and autogenous proliferation.[47]

PATHOGENESIS

Hepatocellular carcinoma has been described as developing from liver cells both with and without cirrhosis, from "rests" within the liver and from adenoma.[48] The transition of adenoma to carcinoma is gradual and adds to the difficulty of differentiating benign adenoma from early carcinoma.[12a] Solid adenoma may also become cystic by accumulation of secretion and develop into cystadenoma, according to Kaufmann [49] and Hall and Brazil.[50] The spread of carcinoma of liver cells depends on the degree of differentia-

40. Steiner, M. M.: Am. J. Dis. Child. **55**:803, 1938.

41. (*a*) Pirie, J. H.: M. J. South Africa **11**:87, 1921. (*b*) Strong, G. F., and Pitts, H. H.: Arch. Int. Med. **46**:105, 1930; Ann. Int. Med. **6**:485, 1932. (*c*) Tull.[3k] Flemming.[16a]

42. Abbe, R., in Dennis, F. S.: System of Surgery, Philadelphia, Lea Brothers & Co., 1896, vol. 4, p. 543. Chaillons, M. J.: Bull. Soc. anat. de Paris **73**:572, 1898. Weidman, F. D.: Proc. Path. Soc. Philadelphia **22**:45, 1919-1920. Rolleston and McNee.[3h] Pepere.[7] Dyrenforth.[11] Pallarés Iranzo and Bartual Vincéns.[33b'] Wright.[33p'] Peck.[34] Gasparian.[35] Schmorl.[37] Dansie.[38] Müller.[39] Steiner.[40] Pirie.[41a] Strong and Pitts.[41b]

43. Yamagiwa.[3m] Karsner.[7g] Abel.[12a] Briggs.[12b] Glynn.[12e] Muir.[12d] Rosenbusch.[12f] Sabourin.[12g] Schiwoff.[12h] Stewart.[12i]

44. (*a*) Rosenthal, S. R.: Tr. Chicago Path. Soc. **14**:17, 1936. (*b*) Rowen, H. S., and Mallory, F. B.: Am. J. Path. **1**:677, 1925. (*c*) Lius.[33w] (*d*) Marino.[33x]

45. Carli, C.: Arch. ital. di chir. **58**:211, 1940.

46. Rhoads, C. P.: Bull. New York Acad. Med. **18**:53, 1942.

47. Goerner, A., and Goerner, M. M.: J. Biol. Chem. **128**:559, 1939.

48. Love, R. J. M.: Brit. J. Surg. **22**:387, 1934. Goldzieher, M., and von Bokay, Z.: Virchows Arch. f. path. Anat. **203**:75, 1911. Trenkel, A.: Zur Pathogenese der Leberadenome: Ein Fall von solitärem Leberzelladenom mit Spontanperforation und Verblutung in die Bauchhöhle, Zurich, H. Rütschi, 1925. Siegenbeek van Heukelom, D. E.: Beitr. z. path. Anat. u. z. allg. Path. **17**:341, 1895.

49. Kaufmann: Zentralbl. f. Gynäk. **31**:913, 1907.

50. Hall, I. W., and Brazil, W. H.: M. Chron. **6**: 243, 1903-1904.

tion, as those of high differentiation spread only within the liver or perhaps to regional lymph nodes at the hilus, while the more anaplastic carcinoma cells survive extrahepatically and are spread by the blood stream to the lungs, bones and even to the spleen.[51]

Characteristic microscopic changes indicative of the degree of malignancy of primary carcinoma of the liver are fairly constant. The adenomatous liver cells are usually larger than those seen in normal liver tissue while the cells of carcinoma are smaller except for the atypical giant cells that are present. Bile secretion and bile plugs are observed in the canaliculi in adenoma but not in carcinoma. With increasing anaplasia the cytoplasm loses its normal granularity and its staining reaction changes from eosinophilic to basophilic. The nuclei become more hyperchromatic and vary in size, with formation of tumor giant cells in highly malignant carcinoma. They often appear to proliferate amitotically. In highly anaplastic carcinoma the cells may no longer appear epithelial. The growth-forming sinusoid keeps pace with liver cell proliferation in benign tumors but does not parallel the growth of carcinomatous cells, so that the latter themselves tend to line the blood sinuses. Liver cells may form pseudo bile ducts, giving the impression of cholangiohepatoma.

Bile duct epithelium also shows characteristic morphologic changes with increased rapidity of growth. There is a loss of duct arrangement, and the cells become variable in size, to produce a papillary, a tubular or a simplex type of carcinoma. The cells become hyperchromatic, rapidly proliferative and invasive.

DIAGNOSIS

A clinical diagnosis of a primary tumor of the liver is possible, and the cancerous type can in many cases be differentiated from the noncancerous on the basis of characteristic symptoms and signs. The pessimism of past reports [52] resulted from observation of only a few cases with contradictory symptoms and signs obscuring certain positive findings common to all. Several authors [53] with extensive experience in these cases have attempted to set forth certain diagnostic criteria [54] enabling early and reasonably certain clinical diagnosis. A large adenoma in the liver is usually easily palpable whether of the cystic or of the solid type. The presence of an intra-abdominal mass in the region of the liver first directs the patient's attention to the lesion. Epigastric fulness or a dragging sensation is more often present than the distinct pain frequently associated with carcinoma. Gastric and duodenal displacement by the mass causes vague indigestion in adults and vomiting in children. Encroachment on the hilus of the liver causes manifestations of portal and biliary obstruction. The diaphragm is occasionally elevated but rarely fixed and then only when necrosis of the adenoma is marked, in contrast to the relative frequency of diaphragmatic fixation in cases of carcinoma.[55] Other manifestations that are rare or absent in cases of adenoma in contrast to their frequent appearance in cases of carcinoma are weakness, loss of weight, anemia, ascites and peripheral edema. Confusion in the diagnosis of adenoma has been caused by erroneous inclusion of two nonadenomatous lesions: the hyperplastic nodules associated with cirrhosis, incorrectly diagnosed as "multiple adenoma" with a clinical picture of cirrhosis, and carcinoma in the form of a single massive growth, mistakenly diagnosed as "malignant adenoma." [22b] The existence of a tumor that has been growing slowly for several months or years before treatment is sought is more indicative of adenoma, for carcinoma has a more rapidly fatal course, death occurring usually within an average of one and one-half to three months.[56]

An analysis of 400 cases of primary carcinoma of liver cells, those with complete histories being chosen from a total of 1,175 [57] published cases,

51. Paterni, L.: Policlinico (sez. med.) **36**:125, 1929.

52. Greene, J. M.: Surg., Gynec. & Obst. **69**:231, 1939.

53. Tull.[3k] Strong and Pitts.[41b]

54. Symmers, cited by Gustafson.[31] Bastianelli.[33d]

55. Benda, R.; Gauthier-Villars, P., and Goulfier, R.: Bull. et mém. Soc. méd. d. hôp. de Paris **56**:449, 1940.

56. Wollstein, M., and Mixsell, H. R.: Arch. Pediat. **36**:268, 1919. Tull.[3k]

57. Acland, T. D., and Dudgeon, L. S.: Lancet **2**: 1310, 1902. de Beule, F.: Rev. belge sc. méd. **3**:232, 1931. Blumenau, E.: Arch. f. Verdauungskr. **27**:1, 1920. Cacciamali, A.: Tumori **9**:377, 1935. Clawson, B. J., and Cabot, V. S.: J. A. M. A. **80**:909, 1923. Cole, W. H.: S. Clin. North America **20**:47, 1940. Coller, cited by Tinker, M. D. B.: Ann. Surg. **102**:728, 1935. Counseller, V. S., and McIndoe, H. H.: Arch. Int. Med. **37**:363, 1926. Cullen, J. S.: J. A. M. A. **44**:1239, 1905. Elliot, A.: Pennsylvania M. J. **1**:193, 1897. Fèvre, M.: Mém. Acad. de chir. **66**:140, 1940. Févre, M., and Dassios, G.: J. de chir. **51**:321, 1938. Finny, C. M., and MacFarlane, L. R. S.: J. Roy. Army M. Corp. **68**:117, 1937. Finucci, V.: Pathologica **23**: 671, 1931. Frankau, C.: Proc. Roy. Soc. Med. (Surg. Sect.) **16**:59, 1922-1923. Freeman, L.: Tr. Am. S. A. **22**:196. Garré, C.: Beitr. z. klin. Chir. **4**:181, 1888; Surg., Gynec. & Obst. **5**:331, 1907. Hamburger, H. S.: Indian J. Pediat. **5**:98, 1938. Hargrove, J. B.: New Orleans M. & S. J. **61**:632, 1908-1909. Hawthorne, G. B.: Brit. M. J. **1**:628, 1901; M. Rev. **4**:265, 1901. Jackson, R. H.: Tr. South. S. & Gynec. A. **44**:299, 1931. Keller, R. M.: Zusammenstellung der während der letzten fünfzig Jahre in der Literatur beschriebenen

(Footnote continued on next page)

and a tabulation of 36 cases of primary carcinoma at the Cincinnati General Hospital yield the results shown in the accompanying table.

Percentage Incidence of Each Symptom of Carcinoma in Three Series of Cases

Symptom of Carcinoma	36 Cases at Cincinnati General Hospital	134 Cases of Tull [28]	400 Cases Reported in Literature
1. Anemia	96	34	68
2. Loss of weight and weakness	94	88	88
3. Palpable tumor	86	68	68
4. Jaundice	80	34	38
5. Pain	71	10	52
6. Fever	53	34	34
7. Vomiting	44	2	12
8. Edema of legs	40	84	60
9. Ascites	37	48	46
10. Fixed diaphragm	18	38	30
11. Tender enlarged liver	15	15	12

The table indicates that the predominant symptoms and signs observed are essentially the same in all studies, but the proportional frequency with which each manifestation occurs varies somewhat. They appear related to (*a*) the proportional difference in type of carcinoma, (i. e., cholangiomatous or hepatomatous), (*b*) to the supposed etiologic basis for the neoplasm as affecting the liver substance (e. g., hepatic infection with flukes as noted in most of Tull's [28] cases reported from the Orient, and alcoholic or dietary deficiency as observed by me) and (*c*) to the general condition of the patient. Tull's [28] patients also appeared to have a nutritional deficiency with edema of the extremities, a common feature, and the loss of weight may have been actually greater and more constant in his than in our cases but hidden by the weight of retained anasarca fluid. Among the cases studied at the Cincinnati General Hospital were 10 of cholangiocarcinoma and 26 of malignant hepatoma. The patients with bile duct carcinoma had an average survival of six weeks to one and one-half months from the onset of symptoms noticeable to the patient. This rapid course appears related to more widespread metastasis and rapid emaciation and cachexia. Sudden death in 2 patients within four days after their admission to the hospital was due to rupture of the tumor and intra-abdominal bleeding. Patients with liver cell carcinoma had an average survival of eight months and were less cachectic and anemic than those with bile duct carcinoma. This difference has been noted by others but not to the same extent. The number of these cases of cholangiocarcinoma is too small to be of statistical value. Checking back on the clinical description of the abdominal mass in comparison with the gross findings at autopsy, I find that palpation is not utilized to the full extent. If intraperitoneal fluid is slight or absent and if the abdominal wall is soft and not too thick, one should be able to determine the size, the shape, the location, the consistency, the movability, the sensitivity and the number of the tumor masses, as stressed by Gatewood.[58] Carcinomatous nodules in the liver are usually hard, irregular, somewhat fixed, multiple and slightly tender in contrast to the more circumscribed, less firm, uniform, movable, single and nontender adenomatous ones. The latter usually project from the anterior edge instead of going deep into the liver substance as do carcinomatous nodules. I found the diaphragm elevated but not fixed in cases of carcinoma, in contrast to the frequency of fixation reported by others.

Secondary carcinoma is considered [4] to differ from primary carcinoma of the liver by evidence of primary carcinoma elsewhere, greater tendency toward multiplicity and diffuse involvement of the liver, slower increase in size, more marked jaundice, ascites and early emaciation in spite of the fact that secondary usually is not as rapidly fatal as primary carcinoma.

Laboratory tests are not of diagnostic importance but are helpful in evaluating the extent of hepatic damage when one is contemplating surgical intervention. Various reports [59] on this subject each claim a different test to be of greatest value, but all are based on determina-

Fälle von Leberadenom mit Berücksichtigung der Beziehungen zur knotigen Hyperplasie und zum Carcinom der Leber, Würzburg, N. Seubert, 1908. Kuznetskiy, D. P.: Vestnik khir. **50**:181, 1937. McArthur, L. L.: Ann. Surg. **42**:626, 1905. MacCallum, W. G.: Textbook of Pathology, ed. 6, Philadelphia, W. B. Saunders Company, 1936, pp. 1054 and 1116. McCreary, J. W.: Pennsylvania M. J. **40**:630, 1937. Mast, W. H., and Streamer, C. W.: J. A. M. A. **100**:1684, 1933. Milne, L. S.: J. Path. & Bact. **13**:248, 1909. Philipp, P. W.: Ztschr. f. Krebsforsch. **5**:326, 1907. Ribbert, H.: Deutsche med. Wchnschr. **35**:1607, 1909. Thöle, F. W. H., in von Bruns, P.: Neue deutsche Chirurgie, Stuttgart, F. Enke, 1913, vol. 6, pp. 51, 101 and 387; vol. 7, p. 304; cited by Tinker, M. B.: Ann. Surg. **102**:728, 1935. Thomson, S. A.: Canad. M. A. J. **32**:675, 1935. Tibiriçá, P. Q. T., and Santos, L.: An. Fac. de med. de São Paulo **7**:133, 1932. Von Glahn, W. C., and Lamb, A. R.: M. Clin. North America **8**:29, 1924. Wegelin, K.: Virchows Arch. f. path. Anat. **179**:95, 1905. Wendel, W.: Arch. f. klin. Chir. **114**:982, 1920. Wheeler, F. J.: Guy's Hosp. Rep. **48**: 225, 1909. White, W. H., in Allbutt, T. C.: System of Medicine, New York, Macmillan & Co., 1897, vol. 4, pp. 194 and 197. Winternitz, M. C.: Johns Hopkins Hosp. Rep. **17**:143, 1916.

58. Gatewood, L. C.: M. Clin. North America **24**: 205, 1940.

59. Bargen, J. A., and Rankin, F. W.: Ann. Surg. **91**:225, 1930. Cohen, P. P., and Hekhius, G. L.: Cancer Research **1**:620, 1941. Meranze, D. R.; Meranze, T., and Rothman, M. M.: Pennsylvania M. J. **41**:1160, 1938. Nadler, W. H., and Wolfer, J. A.: Arch. Int. Med. **44**:700, 1929.

tions of disturbed hepatic function, and disturbance of this function is manifest only when the liver is extensively involved and the lesion inoperable. Abnormally increased prothrombin time and anemia should be corrected preoperatively.

Roentgenograms are useful in localizing the tumor masses, in estimating their size, in determining extension and invasion of the diaphragm or adjacent viscera, in detecting calcification (sometimes indicative of a noncancerous or teratoid tumor), and in ruling out pulmonary or other metastases. A barium sulfate meal will show displacement of the stomach or of the duodenum and will help to exclude unsuspected primary tumor in the gastroenteric tract. A barium sulfate enema, with or without the double contrast by air, will show downward displacement of the colon differing from the forward crowding of renal masses. The making of hepatograms with the aid of colloidal thorium dioxide (thorotrast) is no longer considered advisable because of the damaging effect on healthy liver tissue.

Histologic examination of removed tissue still constitutes the only accurate and reliable method of diagnosing any tumor of the liver. An adequate representative section can best be obtained by exploratory laparotomy and biopsy. This permits visualization of gross characteristics that aid in the recognition of the type of tumor as well as the extent of involvement of the liver, and permits an estimate of the appropriate operative possibilities. Important gross features are: the location of the neoplasm, particularly with relation to hilar structures; the number of tumor masses; the extent of the local involvement, such as fixation to the diaphragm or the neighboring viscera; the presence or the absence of encapsulation; the presence or the absence of regional lymph nodes or of a distant metastasis. Peritoneoscopy offers[60] many of the benefits of laparotomy, including visualization and biopsy. Aspiration biopsy is proposed by the physicians of the Memorial Hospital for the Treatment of Cancer and Allied Diseases, in New York,[61] as a convenient method to get material for microscopic study, the Martin and Ellis method being used. Punch biopsy by Silverman's method is deemed adequate by a few authors.[62]

Of the 7 patients with hepatocellular adenoma observed at the Cincinnati General Hospital, 3 were treated by surgical resection; the patient with the largest tumor was admitted in uremic coma due to nephrosclerosis. In the remaining 3 the tumor was a chance finding at autopsy. In the patients operated on the outstanding clinical features were mild discomfort in the epigastric area and the right upper abdominal quadrant, a slowly increasing mass in the hepatic region, and absence of loss of weight, fever and anemia. Histologically, all of these tumors were composed of uniform aggregates of rather large liver cell cords, the cells having abundant eosinophilic granular cytoplasm and medium-sized central round nuclei showing little hyperchromatism. Rarely, mitotic figures could be seen. Bile ducts and portal triads were absent. The blood supply consisted of normal-appearing sinusoids separating the cords of liver cells. The sinusoids were difficult to demonstrate when the liver cords were cut in cross section, often simulating a mass of liver cells within a blood sinus. These lesions were grossly and microscopically well demarcated, but the thickness of the capsule varied with the rate of growth and the size of the tumor.[3h,1] The large, clinically demonstrable tumors, as in our 4 cases, had thick capsules, but the 3 quiescent smaller ones, of the type often called hamartoma, had delicate or irregular capsules that appeared lacking in a few lesions, such as the one illustrated in figure 1 *B*, though an abrupt change in size and arrangement appears between the normal and the adenomatous tissue.

Variations in adenoma reported in the literature consist of foci of variable amounts of heavily bile-stained cells with intercellular bile plugs, and foci of necrosis that at times become calcified, as well as foci of hemopoiesis.[63] Some larger specimens were described as having heavy interlacing bands of fibrous tissue enclosing irregular nests or broad sheets of liver cells but with the bulk of the cells in columns or in corded arrangement.

As to carcinoma of liver cells, the 26 specimens which I have studied exhibited variation in cell type and staining capacity, loss of polarity, invasion of blood vessels and other evidences of unchecked growth. They showed variable amounts of arrangement into liver cords and lobulation, but this pattern became more indistinct with increase in malignancy. The sinusoidal structures were often reduced to rows of tumor cells lining irregular blood spaces. Proliferative activity was increased largely by atypical mitosis but occasionally by amitosis as well. The cyto-

60. Beling, C. A.: Arch. Surg. **42**:872, 1941. Ruddock, J. C.: South. Surgeon **8**:113, 1939.

61. Binkley, J. S.: Am. J. Cancer **36**:193, 1939. Piaggio-Blanco, R. A., and Paseyro, P.: An. Fac. de med. de Montevideo **23**:964, 1938.

62. Tripoli, C. J., and Fader, D. E.: Am. J. Clin. Path. **11**:516, 1941.

63. Hommerich, K.: Frankfurt. Ztschr. f. Path. **1**: 126, 1907; cited by Ssolowjew, A.: Beitr. z. path. Anat. u. z. allg. Path. **88**:337, 1932.

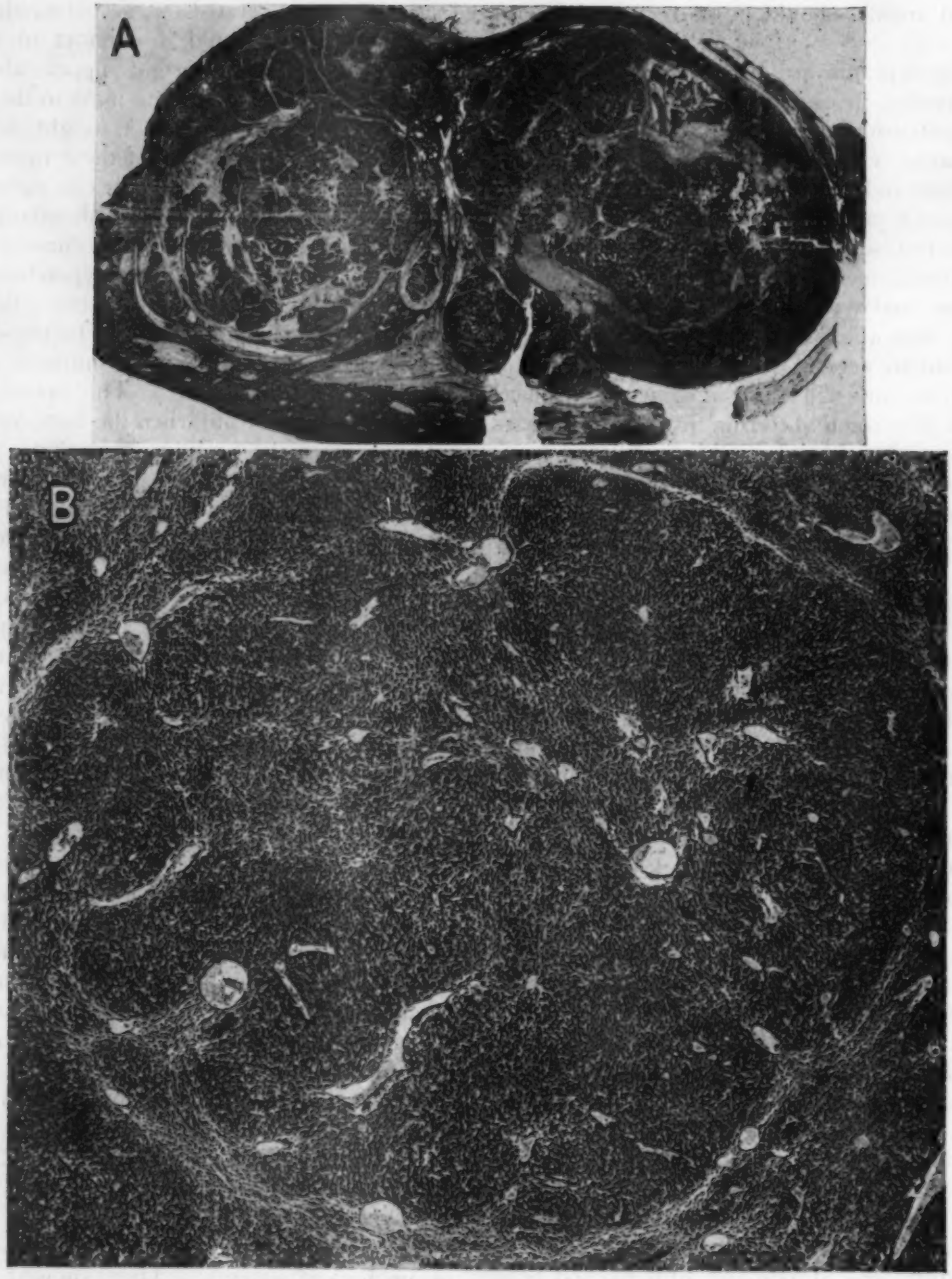

Fig. 1.—*A*, adenoma of liver cells; a large microscopic section of the resected specimen. There is a definite capsule separating the tumor from the normal liver tissue in the lower part of the section. Hematoxylin and eosin stain; × 0.80.

B, adenoma of liver cells in the form of a small quiescent nodule often called the hamartoma type. There is minimal capsule formation but definite demarcation of the tumor, which is composed of liver cells in cords with intervening sinusoids and a normal blood supply but without bile ducts. Hematoxylin and eosin stain: × 16.5.

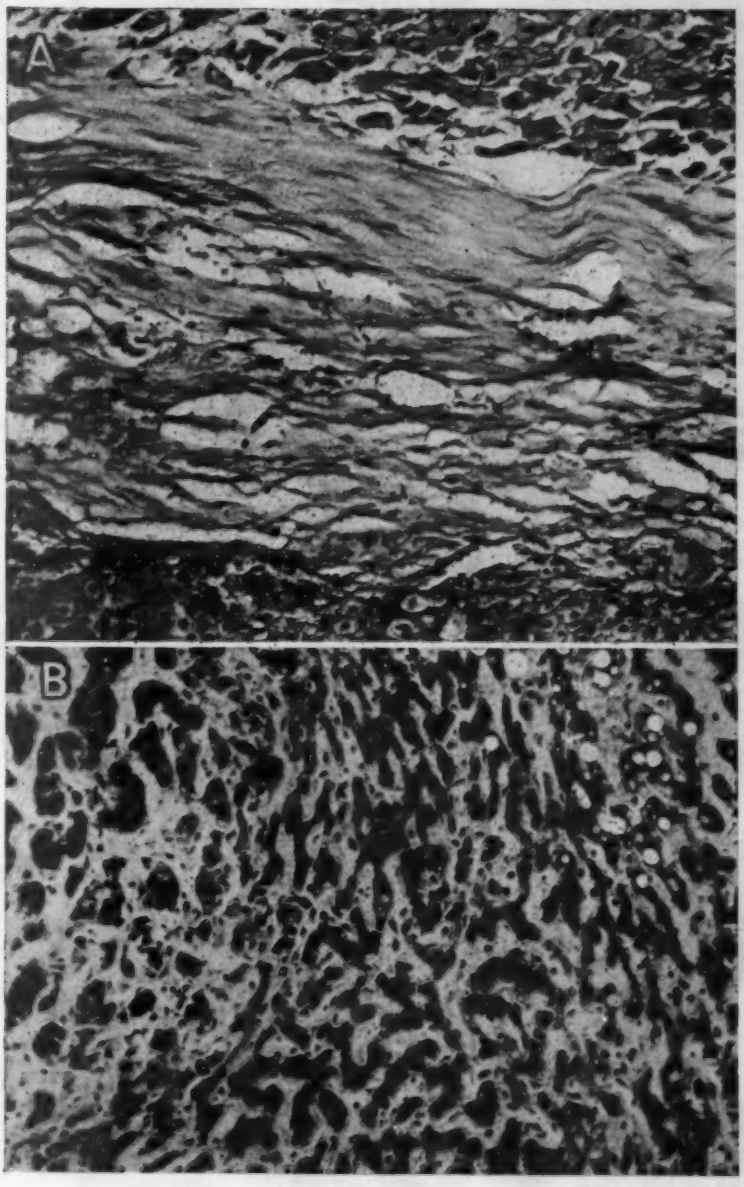

Fig. 2.—*A*, adenoma of liver cells. The thick capsule separates the adenoma above from the normal liver tissue below. Hematoxylin and eosin stain; × 160.

B, carcinoma of liver cells, well differentiated. There is invasion of liver tissue by cells that are clustered rather than arranged in cords. There are reduction of cytoplasm and hyperchromatism of nuclei, some of which are giant in size. Hematoxylin and eosin stain; × 160.

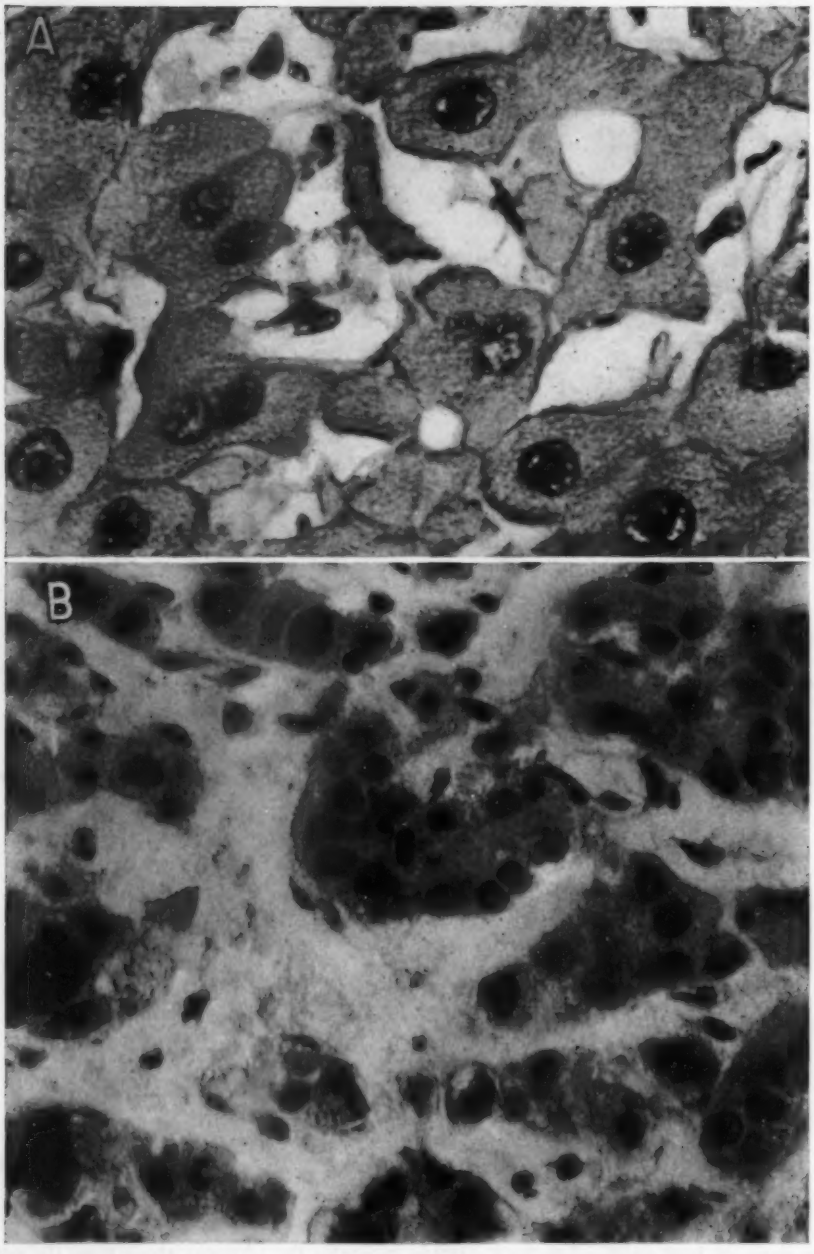

Fig. 3.—*A*, adenoma of liver cells. There is cordlike arrangement of large uniform cells with granular eosinophilic cytoplasm. Hematoxylin and eosin stain; × 650.

B, carcinoma of liver cells, well differentiated. There is retention of some granularity in the cytoplasm of the cells, which are less definitely corded, and the variably sized nuclei are hyperchromatic. There is invasion of normal liver tissue. Hematoxylin and eosin stain; × 650.

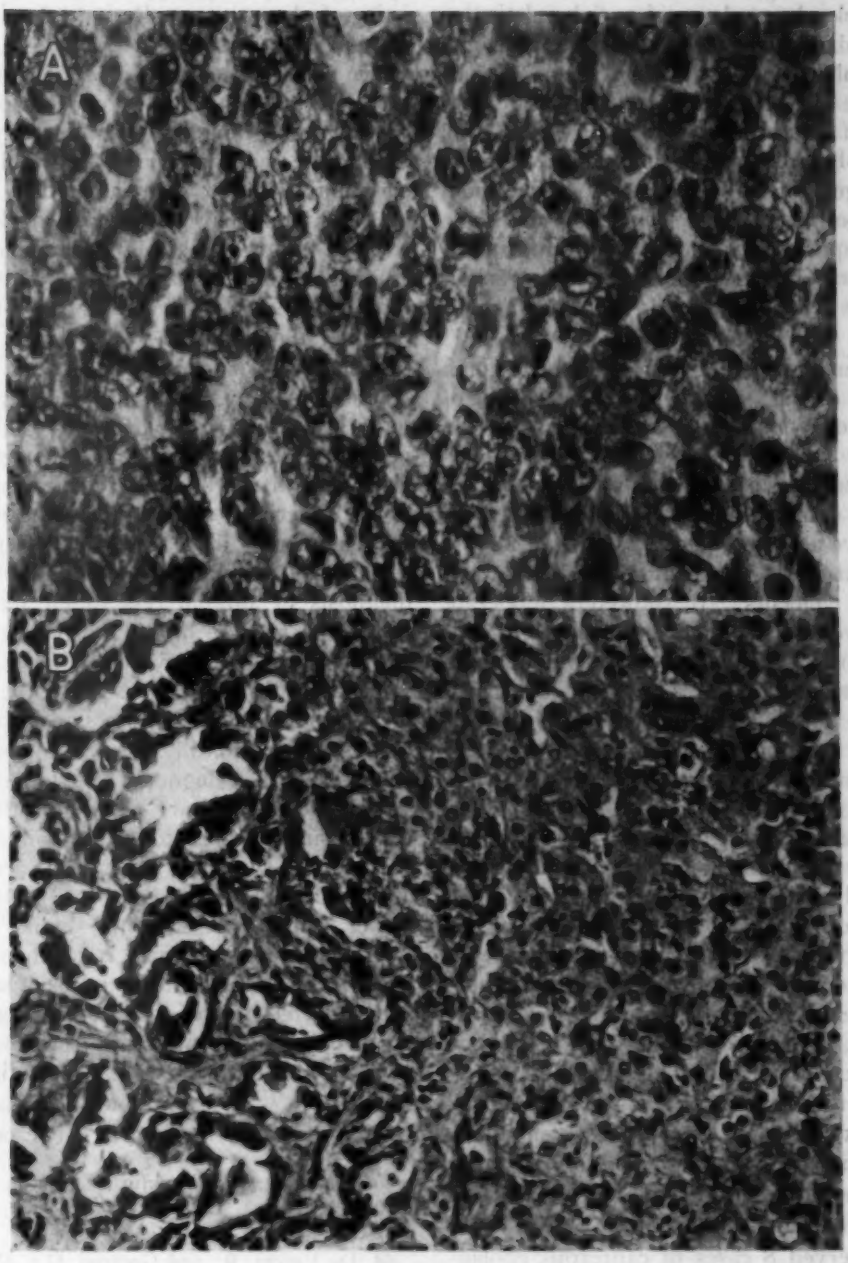

Fig. 4.—*A*, carcinoma of liver cells, highly malignant. There are a loss of the normal arrangement of cells and a marked reduction of cytoplasm, and the variably sized nuclei show numerous mitotic figures. Hematoxylin and eosin stain; × 650.

B, carcinoma of intrahepatic bile ducts. The normal liver tissue is invaded by cells in an alveolar pattern, with hyperchromatic nuclei. Increased fibrous tissue accompanies the tumor growth. Hematoxylin and eosin stain; × 200.

plasm had lost its granularity and had changed from eosinophillic to basophilic in the more malignant type. Regardless of the apparent cancerous or noncancerous appearance of the tumor, any invasion of blood vessels or of peripheral tissues was regarded as evidence of cancerous change. While irregular condensations or bands of connective tissue did lie within some tumors, distinct encapsulation was absent. The uninvolved liver tissue often showed advanced periportal cirrhosis with a suggestion of multicentric origin from atypical hyperplasia; less frequently the liver tissue was quite normal, showing infiltration from a central larger mass, suggesting origin by cancerous change of an adenoma of liver cells.

Only 2 tumors were diagnosed as adenoma of bile ducts of the "solid type," and they were found in autopsy material. Both nodules were encapsulated and composed of small bile ducts lined by single layers of cuboidal epithelium separated by dense strands of fibrous connective tissue. I have also observed at autopsy a nodule which consisted of both hepatomatous and cholangiomatous elements. It showed a moderate number of liver cells containing fat droplets with a central small collection of small bile ducts which was enveloped in a mantle of closely packed small lymphocytes. At the periphery of the liver cells the sinusoids were abruptly dilated, but no capsule was present. There were no recognizable portal triads in the tumor. Solid adenoma of bile ducts is reputed to become cystic because of accumlulation of intrinsic secretion.[64]

I observed a case of cystadenoma of a bile duct in which surgical removal was planned but in which on exposure the neoplasm was found to be too extensive for resection. Microscopically a removed portion showed cystic spaces lined by low, flattened epithelial cells and filled with mucinous material. The epithelial lining of some of the cysts was completely effaced. Other cases reported in the literature show the cyst lining to vary from flattened to columnar epithelium and to contain many layers, at times assuming a papillary arrangement.[65]

I have observed 8 cases of cancerous cholangioma. This type of tumor is distinguished by ductlike structures of cuboidal to tall columnar epithelial cells supported by abundant fibrous connective tissue stroma. The epithelial tumor cells form aggregates in which they appear oval or rounded but never exhibit the delicate interepithelial capillary-supporting stroma characteristic of hepatoma. Periportal cirrhosis was present in 3 of the 8 cases. There was no evidence of secretion of bile in the tumor. Reported variations of specimens of cancerous cholangioma range from medullary carcinoma arising in cystadenoma[66] to highly anaplastic carcinoma simplex characterized by small cords of clear epithelial cells lying in dense fibrous strands. Morphologically, certain forms of carcinoma of the gallbladder are undistinguishable from cholangioma—hence the importance of recognizing the condition of that organ at operation or autopsy.

The mesenchymal tumors primary in the liver need no special pathologic description, as they resemble the tumors of similar origin occurring elsewhere in the body. Hemangioma is usually of the cavernous type; with increasing anaplasia and cellularity it is designated hemangioendothelioma and finally solid endothelioma. The diagnosis of adrenal rest tumor must be made cautiously because of the morphologic similarity of the cells to liver cells that have become vacuolated through deposition of glycogen, fat or hydropic. Some authors feel that until the lipoids are chemically proved to be of the adrenal type the diagnosis should be withheld.[67] In considering primary sarcoma in the liver great care should be taken to exclude highly anaplastic carcinoma, the cells of which are compactly arranged and assume oval or rounded shapes. Other features of sarcoma should be searched for, such as intercellular collagenous substance. Many cases reported as instances of sarcoma have been rejected in later reviews. In some instances the neoplasm may originate from the diaphragm or from the thoracic wall, penetrating the liver secondarily. Fibroids and fibroma also have been reported in the liver.[68] The occurrence of

64. Lenormant, C., and Calvet, J.: J. de chir. **45**:715, 1935.

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primary melanotic tumor of the liver still is doubtful; those who have described such a tumor have attributed its origin to Kupffer cells. If this is the origin, the tumor should be designated as reticuloendothelioma or clasmatocytoma rather than melanoma.

TREATMENT

The treatment of primary tumors of the liver at present is exclusively surgical. Normal and neoplastic liver tissue are both highly radioresistant, and severe systemic reactions resulting from irradiation of the upper part of the abdomen preclude application of radiant energy to these tumors. My colleagues and I have attempted to treat recurrent adenoma (1 case) and carcinoma of liver cells (several cases) with radiation. The treatment had no appreciable effect on the tumors but caused marked nausea and vomiting as well as increasing anemia. This is in accord with the results others have noted with roentgen therapy.[69]

Encapsulated adenoma is the most amenable to surgical removal and can be excised completely with minimal physiologic disturbance.[70] Surgical removal of adenoma and that of solitary circumscribed carcinoma of the liver face the same problem of hemostasis. In recent years this has been accomplished by hemostatic sutures and electrocoagulative cutting. Improved preoperative and postoperative care has prevented fatal metabolic disturbances. Thirty-three cases of resection of solid adenoma of the liver have been reported with immediate postoperative mortality in 9 per cent, recurrence within one to six and one-half years in 21 per cent and with the patient living and well from two and one-half to five years in 70 per cent. We have had 3 cases of resection at this hospital with 1 patient living and well over eight years, 1 living and well over twenty months and 1 with a hilar tumor which could not be completely resected and which recurred after three and one-half years.

Obviously, the tumors diagnosed as liver cell carcinoma are not as amenable to surgical removal, though it seems reasonable that all of these tumors that are circumscribed should be resected if possible, as a rapid fatal termination can be expected if treatment is symptomatic. In addition to the technical feasibility of resection, one must consider the patient's toleration of such an extensive surgical operation. Publications of the past ten years have voiced optimism about attempting surgical removal for carcinoma of liver cells.[71] Some authors[72] still expressed the belief that all attempts at treatment of such tumors of the liver are futile. We have had no cases of resection of frank carcinoma, but perhaps some patients should have been given the benefit of at least laparotomy to determine the operative possibilities. Reports of 40 per cent of patients living and well from three to seven years after resection of carcinoma cannot be ignored in view of the fact that this disease is rapidly fatal in all nontreated ones.[73]

SUMMARY

The classification presented stresses the importance of recognizing that adenoma as well as carcinoma is included in the term "hepatoma." Cholangioma and cholangiohepatoma can also be divided into benign and malignant types. Other primary tumors of the liver are of vascular and stromal origin without specific hepatic elements. Abnormal types of nodular hyperplasia are excluded from true tumors.

The term "liver cell adenoma" is limited to tumors that appear benign in all respects but have a marked potentiality of cancerous transformation. Hamartoma is classified with adenoma. The clinical diagnosis of adenoma is possible because the manifestations of this tumor are local rather than systemic as are those of carcinoma. Adenoma usually forms a palpable mass producing a sensation of pressure or a dull ache in the right upper quadrant of the abdomen, often radiating to the back. Portal or biliary obstruction may occur. Displacement of the gastrointestinal tract causes nausea and vomiting. In contrast to carcinoma, it does not become fixed to the diaphragm, and there is no systemic disturbance as evidenced by loss of weight, weakness, anemia and fever. Laboratory tests show adequate hepatic function. The roentgenogram reveals a localized mass in the liver which may exhibit calcification and no evidence of metastasis. Peritoneoscopy confirms the absence of intra-abdominal extension. Histologically, the hepatomatous adenoma is composed of uniform aggregates of large liver cells in cords. The cells have abundant eosinophilic granular cytoplasm and medium-sized central round nuclei showing little hyperchromatism. Rarely, mitotic

69. Pohle, E. A.: Clinical Roentgen Therapy, Philadelphia, Lea & Febiger, 1938, p. 231. McRae.[6c]

70. (*a*) Adler, H.: Zentralbl. f. Chir. **63**:987, 1936. (*b*) Anschütz, W.: Samml. klin. Vortr., 1903, no. 356-357 (Chir. no. 99), p. 451. (*c*) Coryn, G.: J. de chir. et ann. Soc. belge de chir. **35-33**:357, 1936. (*d*) Czirer, L.: Magyar orvosi Arch. **25**:157, 1924; abstracted, Surg., Gynec. & Obst. **41**:27, 1925.

71. Keen, W. W.: Boston M. & S. J. **126**:405, 1892. Thomson, J. E.: Ann. Surg. **30**:284, 1897. Yeomans, F. C.: J. A. M. A. **64**:1301, 1915.

72. Boyce, F. F., and McFetridge, E. M.: Internat. S. Digest **18**:67, 1934. Lilienthal.[33v] Johansson.[33s] Rhoads.[46]

73. Rowen and Mallory.[44b] Carli.[45]

figures can be seen. Bile ducts and portal triads are absent. Sinusoids, apparently normal, separate the cords of liver cells. The tumor is demarcated by a definite capsule which varies in thickness in proportion to the size of the tumor. Evidence of secretion of bile and areas of calcification are often present. The treatment consists in surgical removal. The prognosis is good.

Bile duct adenoma has similar clinical diagnostic features except that the more common cystic type tends to be larger and grows more rapidly. All are encapsulated and may be lobulated. Microscopically, the tumor cells are flattened to columnar bile duct epithelial cells and are arranged in solid strands or form cystic spaces. The cysts contain mucinous material, and their lining is flattened or in papillary formation. Surgical excision is the treatment of choice when possible, but marsupialization is safer for a very large tumor of this type.

Carcinoma of liver cells is the most common primary tumor of the liver. The clinical diagnostic features are those of local invasion and systemic disturbances. Fixation to the diaphragm and surrounding viscera is common. Anemia, loss of weight, weakness, jaundice, fever and ascites are often present. Laboratory and roentgenologic studies indicate extensive destruction of the liver and distant metastasis. Histologically the tumor is characterized by unchecked growth with invasion of blood vessels. The normal hepatic architecture is lost and irregular blood spaces lined by tumor cells replace the sinusoids. The cells evidence increasing anaplasia with variation in size and loss of polarity. The cytoplasm is basophilic and nongranular and lacks signs of secretion of bile. The hyperchromatic nuclei show proliferative activity by mitosis or amitosis. The treatment consists in surgical removal except when the involvement of the liver is too extensive or when metastasis has occurred. The number of successful surgical excisions reported is encouraging.

Malignant cholangioma has clinical features resembling those of malignant hepatoma except for a more rapid course reducing the average survival after the onset of symptoms to one and one-half months, compared with the average eight month survival of the patient with carcinoma of liver cells. Histologically the carcinoma of bile ducts consists of cuboidal to tall columnar clear epithelial cells supported by a dense fibrous stroma. It does not have the delicate capillary-supporting stroma characteristic of hepatoma. It resembles certain forms of carcinoma of the gallbladder; so the condition of that organ must be known.

Primary tumors of the liver not containing specific hepatic elements are largely of angiomatous origin. They vary from simple hemangioma to sarcomatous angioma or endothelioma. Numerous reports of fibroma and fibrosarcoma have been published. Rare cases of adrenal rest tumor and 3 probable cases of primary melanoma of the liver were found in the literature. There is no uniformity in the treatment of these tumors.

FEATHER GERM REACTION TO URINE FROM PATIENTS WITH CANCER AND OTHER CONDITIONS

A PRELIMINARY STUDY

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The use of the feather germ of the Brown Leghorn male as a possible indicator of tumorous conditions was suggested by an interesting observation of Greenwood,[1] who found a development of hen-feathering in a capon of this breed in the presence of a tumor about halfway down one kidney.

Greenwood believed that this growth elaborated some substance which caused the assumption of the female feathering, and while his hypothesis was not put to test by grafting fragments of this tumor to other fowl, it nevertheless remains most suggestive, especially in view of more recent chemical studies which indicate certain relations between estrogens and carcinogens (Fieser [2]; Owen [3]).

It might be thought that in the tumors or in the blood stream or the urine of persons harboring tumors substances might be found which when recovered and injected in appropriate dosages into Brown Leghorn cocks and capons would call forth feather changes comparable to or identical with female feathering. If these cocks and capons showed plumage changes of fairly consistent nature, comparably induced feather modifications might to some extent prove serviceable as indications of tumorous conditions suspected to be present in patients. For the purpose of repeated tests with regard to a given patient, the urine offers definite advantages as a source. In a first exploratory approach, therefore, the plumage reactions of Brown Leghorn capons and cocks to urines from known cancerous patients, to urines from persons with other pathologic conditions and to urines from normal persons were recorded.

The greater part of the urines tested were obtained through the cooperation of members of the staff of the Memorial Hospital of the University of Maryland, at Baltimore.

Dr. H. R. Bird of this department gave advice on the treatment of samples.

PROCEDURES

Samples of urine were collected from patients with different cancerous conditions as they presented themselves, and whenever possible twenty-four hour specimens were obtained. As controls, urines were obtained from persons with pathologic but noncancerous conditions and from normal young men. So far as convenient it was planned not to include urines of younger women in this preliminary study since the estrogens might prove a complicating factor and since it had been shown that readily tolerated doses of native urine from pregnant women will induce female feathering in the Brown Leghorn capon (Juhn, D'Amour and Gustavson [4]).

All samples were placed in an ice box on collection, transferred to thermos jars for transportation to the laboratory and again placed in an ice box.

Each sample was tested as soon after receipt as possible. It was first concentrated in vacuo ten to twenty fold over a water bath, the temperature of which was kept around 45 to 50 C. and not allowed to surpass 55 C. In the latter part of the experimental series test capons were not immediately available. The samples of urine collected at that time were treated as follows: Two hundred cubic centimeters of the urine was reduced to 10 cc., to which 0.5 cc. of toluene was added and the mixture shaken. The mixture was placed in a small vial, 1.5 cc. of toluene was layered on the surface and the vial was then placed in the ice box. For use the toluene was driven off by steam and the sample cooled just previous to injection.

The methods developed by Juhn and Gustavson [5] in their use of the feather germ as an indi-

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1. Greenwood, A. W.: Proc. Internat. Cong. Genetics **2**:266, 1932.

2. Fieser, L. F.: Am. J. Cancer **34**:37, 1938.

3. Owen, S.: Quart. Rev. Biol. **12**:340, 1937.

4. Juhn, M.; D'Amour, F. E., and Gustavson, R. G.: Endocrinology **14**:349, 1930.

5. Juhn, M., and Gustavson, R. G.: Proc. Soc. Exper. Biol. & Med. **27**:747, 1930.

cator of estrogen were adopted without change in the present experiments: Regions in the breast tracts of the male are given a preliminary plucking, and when the regenerates attain a suitable length, control preparations are made just before injection of the substance to be tested. For these preparations feather germs are removed, slit lengthwise along one prospective vane-half, flattened gently with the pulp in place between two slides and fixed in 95 per cent alcohol for twenty-four hours or more. On removal of the pressure the feather germs remain affixed to one slide by the pulp, the sheath side forming the free surface. The preparation is then shaken free of alcohol and flooded with a mounting medium (euparal yellow), and a cover slip is added. Clearing takes place in the euparal, and the mounts are permanent.

Pigment modifications may be induced only in the basal region of the feather germ, in a zone which becomes continually displaced in the apical direction with growth of the feather. Preparations made at successive intervals thus record first the effect of a given injected substance and then the time of its effective persistence after administration is discontinued.

Injections of theelin cause female salmon pigment to be laid down in the base of the feather germ in brilliant contrast to the already formed black male tips.

In the tests of the samples of urine a similar modification of pigment but to a deeper, rusty red was considered a positive reaction.

The urine concentrate to be tested was given in subcutaneous injection on each of two successive days. As controls for the normal black color of the male feather germ, preparations of representative samples were made just before the first injection of test urine was given. Preparations were made again three to four days later, and pigment modifications to red in the basal zone of the feather germs were obtained when the response was positive. Preparations made five or six days later showed a displacement of the modified red segment and a reversion to the normal black coloring of the basal zone. This illustrated the limited effective time in the organism of the substances injected.

The larger number of the urines proved toxic to the birds; severe local disturbances developed at the sites of injection which were reminiscent of effects caused by early unpurified sex hormone extracts. Attempts were made to reduce the local disturbances by adjustment of the p_H of the urines to neutrality, but this procedure was ineffective, and the losses among the experimental birds depleted the stock.

INITIAL EXPERIMENTS

In preliminary experiments untreated urines were tested. These were obtained from 2 normal males and 1 normal female and from 2 males with cancer of the liver and 1 female with cancer of the cervix and were given in 20 cc. amounts in the early morning and in the same amounts again in the late evening. The results all proved negative although 40 cc. of urine from a pregnant woman had been demonstrated to cause hen-feathering.[4] However, the cancerous urines gave a positive reaction when they were injected in equivalent amounts but in tenfold concentration; the normal urines similarly concentrated and given in heavier dosage continued to give negative results.

In further tests the urines were all concentrated approximately tenfold and the equivalents of 20 cc. of the fresh urines were injected subcutaneously on two successive days. As regards urines from the male patients, positive reactions were obtained with urines from patients with carcinoma of the rectum, squamous cell carcinoma, infected ranula (the patient had a previous history of carcinoma) and noncancerous gastric ulcer. A negative result was given by the urine of a male whose disease was suspected of being carcinoma but was later diagnosed as noncancerous. Among these samples, the urine from a male with cancer of the rectum proved distinctly less toxic, and so some additional tests were made with it: First, a Brown Leghorn capon, K-19, was given a daily injection of 2.0 cc. of the urine concentrate for seven successive days. The saddle and hackle feathers became modified in the female direction; the condition of the head furnishings, which was also recorded, remained unchanged.

In a second experiment 1 male and 1 female chick, each 15 days of age, were given daily injections of 0.25 cc. of this same urine concentrate for eight days and then were killed and examined. The female chick showed stimulation of the ovary and the oviduct, verified by microscopic investigation; the male chick appeared unaffected. In this case of carcinoma of the rectum the urine apparently contained estrogenic and gonadotropic substances but no androgen as there was no stimulation of the comb, which, as is well known, serves in the so-called capon test.

Of urines from female patients, samples from 2 women who had undergone operative removal of adenocarcinoma of the uterus and carcinoma of the anus respectively gave negative reactions. A positive reaction was found for a young woman (aged 27) with extensive pulmonary tuberculosis and in 1 case of tubal pregnancy.

A final series included a larger number of patients and covered a wider range of pathologic conditions. In order to give as comprehensive a survey as possible in view of the diversity of the test cases, the following grouping was made:

TESTS WITH URINES FROM FEMALES

Positive reactions were observed with urines from patients with the following conditions, including those to whom roentgen or radium treatment was being given: cancer of the breast, 6; cancer of the uterus or the cervix, 13; cancer of the ovaries, 2; cancer of the rectum, 1; generalized abdominal carcinomatosis, 1; osteosarcoma

of the skull, 1. A positive reaction was given with the urine of 1 patient who was suspected of having a cancer in the abdomen.

Negative reactions were obtained after surgical intervention in 3 cases of cancer of the breast, 3 cases of cancer of the uterus and 1 case of tumor of the brain. Negative reactions were found in 1 case of suspected cancer of the stomach and 1 case of suspected cancer of the head of the pancreas.

As regards cases with diagnoses other than cancer, positive reactions were obtained in 1 instance of perforating duodenal ulcer which had caused an abscess in the abdomen and 1 instance of tuberculous adenitis.

In a number of cases included in the foregoing summary it was possible to obtain samples of urine at intervals from the same patient, and the results with these are of interest: (*a*) Woman 72 years old with cancer of the cervix. First test positive; test two weeks later positive. (*b*) Woman 45 years old with cancer of the ovaries. First test positive; test seven weeks later positive. (*c*) Woman 52 years old with carcinoma of the cervix under radium therapy. First test positive; test one week later positive. (*d*) Woman 63 years old with carcinoma of the cervix under radium therapy. First test positive; test three weeks later negative. (*e*) Woman 75 years old with cancer of the breast under roentgen treatment for the past two weeks. First test positive; second test, three weeks later, positive; third test, two and a half months later and eighteen days after surgical removal of growth, negative. (*f*) Woman 63 years old with carcinoma of the uterus who had undergone hysterectomy and salpingo-oophorectomy. First postoperative test negative; test seven weeks later positive. (*g*) Woman 37 years old with tumor of the brain. First test negative; test after partial surgical removal, three weeks later, positive.

TESTS WITH URINES FROM MALES

A positive reaction was noted with urines from patients with a clinical diagnosis of cancer in 1 case each of sarcoma of the knee, carcinoma of the colon, cerebral tumor, retroperitoneal sarcoma, carcinoma of the scalp after partial surgical removal, cancer of the rectum, metastatic carcinoma of the right side of the neck, carcinoma of the bladder and a history of fibrosarcoma.

Repeat tests were made in 2 cases, neither included in the foregoing summary. One was a case of carcinoma of the stomach. The original test was positive. The repeat test, three months later and seven days after surgical removal of the growth, was negative. The second instance was one of carcinoma of the pancreas and massive metastases to the mesentery of the small intestine and metastatic carcinoma of the liver. The first test was positive. The repeat test, three weeks later, was positive.

Positive reactions were observed in a case with the diagnosis of possible Hodgkin's disease and in another in which the diagnosis was probable carcinoma of the stomach.

A negative reaction was observed with urines from persons with a clinical diagnosis of cancer in 1 case each of cancer of the cecum, carcinoma of the larynx under roentgen treatment, carcinoma of the prostate gland and other diseases, cancer of the face under roentgen treatment, carcinoma of the stomach, primary carcinoma of the lung, abdominal carcinomatosis, bronchogenic carcinoma and recurrent basal cell carcinoma of the skin.

Negative reactions were obtained in single cases in which the following cancers were suspected: probable lymphosarcoma of the mediastinum, receiving radiation; probable carcinoma of the large bowel; probable carcinoma of the stomach; probable carcinoma of the head of the pancreas.

Negative reactions were obtained with urines from 6 patients after surgical intervention for cancer, from 4 persons with disease other than cancer and from 6 persons without disease.

A positive reaction was noted in a case of gastric ulcer, and another may have been obtained in an instance of syphilis but the toxicity of the sample in this instance made a determination difficult.

COMMENT

The experimental limits of the tests described set the frame within which conclusions may be drawn as to the possible role of the feather germ as an indicator of cancer. In these tests the negative results with the urines of surgically treated, noncancerous and normal persons are considered as controls.

In the female group the organs affected are in the greater number of cases those manifesting the primary and the secondary sex characters, and there is good agreement between the reaction in the feather germ and the clinical evidence. Since it might be thought that some substance effective in modifying feather pigmentation is normal to the age group represented, known ages of all the women tested were tabulated in relation to negative or to positive reactions. The overlap in distribution proved such that the age factor does not appear to enter into the reaction.

In the wider age range of the male group there was a similar distribution of positive and negative reactions at all age levels. In this group the incidence of cancer was more varied, and a proportionately larger number of negative reactions was noted. In comparing positive and negative tests it was found that in all of the cases of cancer of the liver and cancer of the rectum the tests were positive and that only one location of cancer, the stomach, was associated with both positive and negative reactions. The numbers are of course too small to be other than suggestive; the difference in reaction might be thought of as reflecting an initial difference of the tumors but this could be verified only by extended investigation. It seems probable, however, that in cases of cutaneous cancer pigmentary modifications of the feather germ are not obtained.

In regard to other pathologic conditions, the positive response in 2 cases of gastric ulcer was mentioned.

It is possible that the application of chemical methods would serve to increase the specificity of the reaction although it is recognized that with purification certain biologic effects may be

lost. There is some evidence that the substance or substances which will modify feather pigmentation may be inactivated or destroyed by prolonged exposure of the urines to water bath temperatures of less than 50 C. or with a small distilling surface. Possibly extended sojourn at room temperatures may have a similar effect. Simple detoxification, however, would make longer periods of treatment possible, leading in turn to biologic identification of the substances excreted under given conditions; the adult Brown Leghorn male recording as shown here the presence of estrogens and serving as a subject of test for the androgens, with the chick acting as a subject of test for the gonadotropins.

The toxic attributes of the greater number of the urines might conceivably participate in the pigmentary reaction of the feather germ. To check this point, the local effects noted for each sample were set against the response in the feather germ for every cock and every capon used in the tests. When, as in a number of instances, a fowl served repeatedly, this tabulation also measured individual variability to some extent. No relation was found between the degree of local disturbance caused by any one sample and its effect in the feather germ.

A final point deserves mention. The Brown Leghorns used in most of the later tests proved to be of a strain with a relatively high degree of susceptibility to the avian leukosis complex; this suggested a comparison of the records of test fowl in which at some time after cessation of experiments symptoms of the disease developed with the records of test fowl apparently free. This comparison showed that samples positive in one group were also positive in the other; samples negative in one group were likewise negative in the other. It is nevertheless possible that the sensitivity of an entire susceptible strain may be of a different level in comparison with that of a resistant one.

SUMMARY

The pigmentation reaction of the growing feather germ of the Brown Leghorn capon has been used to record the effects of injected urines from cancerous, other pathologic and normal subjects. With the injection of the concentrate equivalent of 20 cc. of fresh urine on two successive days, the responses obtained from urines of the main classifications, including some obtained on repeated tests, were as follows:

From urines of males: Cancer—15 positive reactions in 24 tests. Cancer suspected—2 positive reactions in 6 tests. After surgical intervention for cancer—7 negative and no positive responses. Other conditions—3 positive reactions in 8 tests. Normal—8 negative and no positive reactions.

From urines of females: Cancer—27 positive responses in 28 tests. Cancer suspected—1 positive reaction in 3 tests. After surgical intervention for cancer—9 negative and 1 positive reaction. Other conditions—3 positive reactions in 3 tests.

Case Reports

A MIXED TUMOR OF THE SALIVARY GLAND TYPE ON THE LEFT HAND

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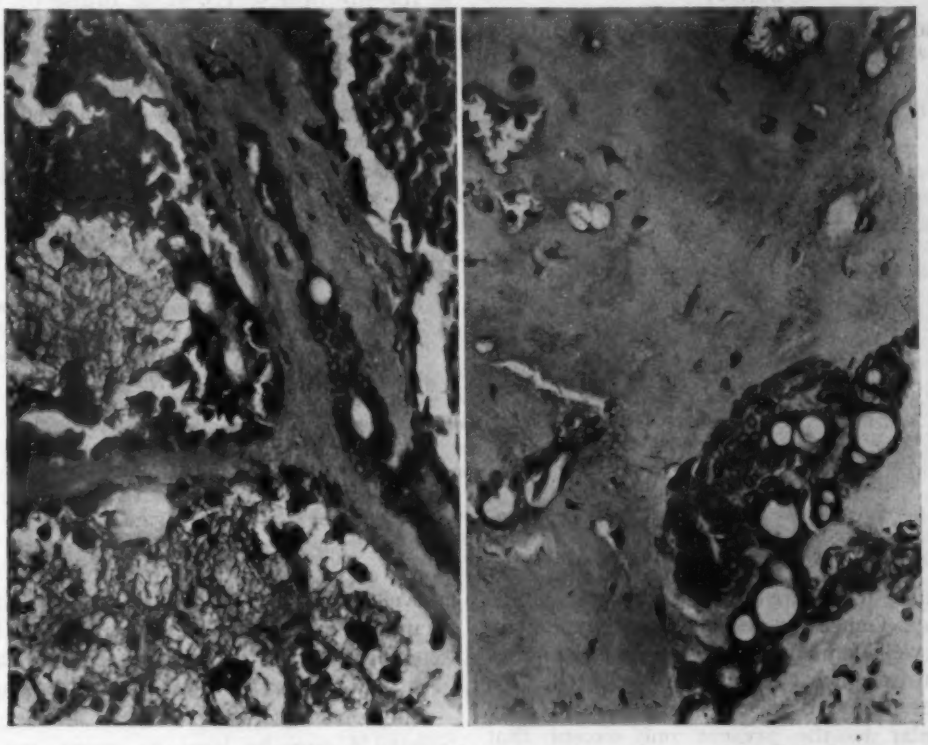
Mixed tumors of the salivary gland type occurring elsewhere than on the head and neck have rarely been reported. Accordingly, the reporting of an additional one appears justified.

A tumor the size of a pecan was removed from the lateral surface of the left hand at the base of the little finger of an 80 year old Indian man. It had been growing slowly over a period of about five years, had produced no symptoms and was thought to be a sebaceous cyst. The clinical data were supplied by Dr. G. D. Waller, Claremore, Okla. Grossly, the formaldehyde-fixed specimen was a firm gray ovoid mass, about 30 by 25 by 20 mm. in diameter, with a slightly scalloped smooth outer surface. The cut surface was mottled grayish white and showed a few pinpoint dark hemorrhagic areas.

Microscopically, the specimen showed a thin, focally incomplete capsule of sparsely cellular dense fibrous tissue. A few trabeculae extended from this capsule into the interior and divided the tumor into ill defined lobules. These showed numerous irregularly shaped sheets, nests, strands and occasional acini of pleomorphic epithelial cells. Most commonly, the cells were polyhedral or fusiform and medium in size, and showed fairly distinct outlines, a moderate amount of lightly to deeply eosinophilic cytoplasm and round to oval nuclei with fine to coarse chromatin granules. Rare mitoses were seen. Less commonly the cells resembled squamous cells of the stratum mucosum, but no intercellular bridges, keratohyaline granules or pearls were clearly seen. In a few masses the cells appeared similar to those of basal cell epithelioma. Occasional cells were large and either were binucleated or showed a single large nucleus.

The arrangement of the cells varied, was often quite irregular and rarely was concentric. The cells at the margins of some masses were fusiform and arranged in slender diverging strands which blended with the surrounding stroma. In some areas, many of the cells were isolated or lay in small groups separated by a small to moderate amount of eosinophilic hyaline or



Left: Area of tumor showing epithelial masses, acini, myxomatous tissue and hyalinized stroma. Romanovsky stain. × 205.

Right: Area of tumor showing acini, myxomatous tissue and border of a cartilaginous zone. Romanovsky stain. × 205.

From the Pathology Laboratory, National Institute of Health.

metachromatically basophilic fibrillar mucoid material. They were stellate or more commonly rounded, with an eccentric nucleus. A few such areas of myxomatous tissue graded into small areas of cartilage.

Acini occurred singly and in small groups apart from, within or continuous with the epithelial masses. They varied from about 25 to 100 microns in diameter, were lined by one to several layers of flattened to cuboidal or polyhedral cells and were either empty or contained some lightly to deeply eosinophilic hyaline material. A few were cystic; the largest was about 2 mm. wide. The fibrous stroma between epithelial masses varied in amount, density and cellularity, was focally fatty and showed extensive hyalinization, focal basophilic degeneration, a few scattered small hemorrhagic extravasations and some irregularly scattered lymphocytes and hemosiderin-laden phagocytes.

COMMENT

The findings in similar cases gathered from the literature [1] have been recorded in the accompanying table. An additional tumor on the hand reported by Cid [2] in 1940 is not listed because I was unable to secure any report with details of the case. Generally, these tumors were firm, lobulated, well defined, round or oval subcutaneous structures which were easily enucleated. No recurrences were recorded. Histologically, they were similar to the present one except that some showed prickle cells and pearl formation. Scharla's [1f] tumor contained no cartilage.

Mixed Tumors of the Salivary Gland Type Reported in the Literature Occurring Elsewhere Than on the Head and Neck

Author	Year	Age	Sex	Duration, Years	Size	Location
Tessmann [1a]	1911	Middle aged	F	4	Larger than cherry	Back of hand near wrist
Kreibig [1b]	1931	38	M	6	Hen's egg	Anterior part of tibia (R)
Kreibig [1b]	1931	30	M	3	Hazelnut	Ventral surface of forearm (L)
Hirsch [1c]	1933	34	F	8	75 by 55 by 45 mm.	Thigh (L)
Gaehtgens [1d]	1934	66	F	2	Cherry	Fourth finger (R)
Vidari [1e]	1935	58	M	Many	Cherry	Calf (L)
Scharla [1f]	1936	30	F	?	Hazelnut	Fifth finger (R)
Simard [1g]	1938	76	F	10	50 by 45 by 42 mm.	Hypothenar region (L)
Uggeri [1h]	1939	29	M	1½	Small nodule	Middle finger (L)
Hollander [1i]	1940	40	M	3	Chestnut	Fifth finger (R)

Many theories have been proposed to explain the origin of these tumors. I am inclined to favor the view of Ewing [3] and others who believe that these tumors are essentially epithelial and that the stromal portions, particularly the cartilaginous and myxomatous tissues, are epithelial products. Thus, there were seen in some areas of the tumor observed by me gradual transitions from compact epithelial masses to loose myxomatous tissue and from typical epithelial cells to chondrocytes.

It has been proposed [4] that such a transformation from epithelium to cartilage might be favored by necrosis, hemorrhage, edema and inflammation subsequent to trauma, stress and strain leading to a dispersal of the cells with the formation of a mucinous and cartilaginous matrix. In connection with this belief it is interesting to note that the tumor in Kreibig's first case occurred at the site of a severe stone bruise on the tibia, that the tumor on the thigh in Hirsch's case was subjected to repeated injury, that the present tumor showed some hemorrhage and hemosiderin deposits and that in most of the recorded cases as well as in the one described here the tumor occurred on the hand, where doubtless there was exposure to repeated trauma. These facts suggest that trauma may be a predisposing factor.

The source of the epithelium, as suggested by Simard,[1g] may be local in origin, possibly, in view of the aciniform structures, derived from sweat glands.

SUMMARY

A mixed tumor of the salivary gland type occurred on the lateral surface of the left hand of an 80 year old Indian man. Ten similar tumors occurring elsewhere than on the head and neck were collected from the literature. All of them were on the extremities, 6 being on the hands. Trauma is suggested as a possible predisposing factor. The view is advanced that these tumors are essentially epithelial in origin, possibly derived from sweat glands, and that the stromal portions, particularly the cartilaginous and myxomatous tissues, are epithelial products.

1. (*a*) Tessmann, E.: Ueber eine Mischgeschwulst in der Gegend des Handgelenks von histologischen Charakter der Mischtumoren der Speicheldrüsen, Inaug. Dissert., Würzburg, C. J. Becker, 1911. (*b*) Kreibig, W.: Frankfurt. Ztschr. f. Path. **42**:281, 1931. (*c*) Hirsch, E. F.: Arch. Path. **16**:494, 1933. (*d*) Gaehtgens, G.: Frankfurt. Ztschr. f. Path. **47**:374, 1934. (*e*) Vidari, E.: ibid. **48**:209, 1935. (*f*) Scharla, O.: ibid. **49**:269, 1936. (*g*) Simard, L. C.: Am. J. Cancer **33**:182, 1938. (*h*) Uggeri, C.: Gazz. d. osp. **60**:848, 1939. (*i*) Hollander, F.: Zentralbl. f. allg. Path. u. path. Anat. **75**:145, 1940.

2. Cid, J. M.: An. de cir. **6**:374, 1940.

3. Ewing, J.: Neoplastic Diseases, ed. 4, Philadelphia, W. B. Saunders Company, 1940, p. 790.

4. Allen, A. C.: Arch. Path. **29**:589, 1940.

MYASTHENIA GRAVIS

Report of a Case with Autopsy

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The genesis of myasthenia gravis and particularly the role of the thymus in it are not clearly understood. It is therefore desirable that all cases fully studied should be reported. In this paper we describe a case of myasthenia gravis with complete autopsy.

REPORT OF CASE

M. L., a 59 year old white man, was in his usual health until four weeks previous to his admission to Mount Sinai Hospital, in Cleveland, July 27, 1941. At that time he became conscious of inability to open his right eye fully. About one week later he began to notice dyspnea on walking and dysphagia such that the only food he was able to swallow was liquids and soft solids. More solid food gave him the sensation that it was "sticking half way down." Next he noticed that after speaking for three to four minutes his speech became thickened and blurred. His most distressing symptom was dyspnea and wheezing on the slightest exertion.

The physician whom he consulted considered a mediastinal tumor on the basis of the progressive dysphagia and dyspnea. A roentgenogram revealed two tumor masses; the first appeared as a dense homogeneous shadow occupying almost the entire left supraclavicular space; the second, as a large lobulated shadow in the anterior mediastinum overlying the arch of the aorta and the base of the heart. The roentgen diagnosis was primary tumor of the apex of the left lung with secondary involvement of the hilus. The patient was admitted to the hospital for bronchoscopic and bronchographic studies.

He was a well developed, well nourished adult, who appeared chronically ill. His temperature, pulse and respiration were normal. There were marked ptosis of the right eyelid, questionable drooping of the left side of the mouth on smiling, inability to wrinkle the forehead, protrusion of the tongue to the left on opening the mouth and many flat and raised, darkly pigmented warts on the trunk.

In an attempt to make a bronchoscopic examination paralysis of the abductor muscles was noted, and after neurologic consultation the diagnosis of myasthenia gravis with tumor of the thymus was made. Ten minutes after an injection of a solution of neostigmine methylsulfate (1 ampule, 1:2,000) the patient showed marked clinical improvement. Roentgen irradiation of the mediastinum was started, and a total of 1,260 r was given in six divided treatments. The administration of neostigmine was continued, at first intramuscularly, later by mouth, and the patient showed marked symptomatic improvement.

The blood counts revealed 11,000 white and 4,479,000 red cells per cubic millimeter; the hemoglobin content was estimated at 85 per cent; the blood sugar amounted to 93 mg., the nonprotein nitrogen to 63 mg. and creatinine to 1.6 mg. per hundred cubic centimeters; the Kline test was negative.

From the Department of Laboratories, Mount Sinai Hospital.

The patient was discharged on August 7 but was readmitted twice because of difficulties in expectorating phlegm. His daily doses of neostigmine had to be gradually increased to 10 tablets of 15 mg. of neostigmine bromide each. During the third hospitalization he had several relapses and was given a solution of neostigmine methylsulfate intramuscularly. After an infusion of 200 cc. of 10 per cent dextrose in saline solution, he suddenly became dyspneic and was unable to talk. In spite of an additional intramuscular injection of neostigmine methylsulfate, he died twenty minutes later.

Autopsy (ten hours after death).—The skin had a peculiar yellowish gray hue. In the anterior mediastinum an encapsulated soft spherical tumor was found, weighing 55 Gm. and measuring 8 cm. in diameter. It could be removed easily by blunt dissection. The surface was even; the upper pole felt cystic. The cut surface was grayish pink and had a honeycombed appearance. Delicate fibrous strands traversed the tumor irregularly.

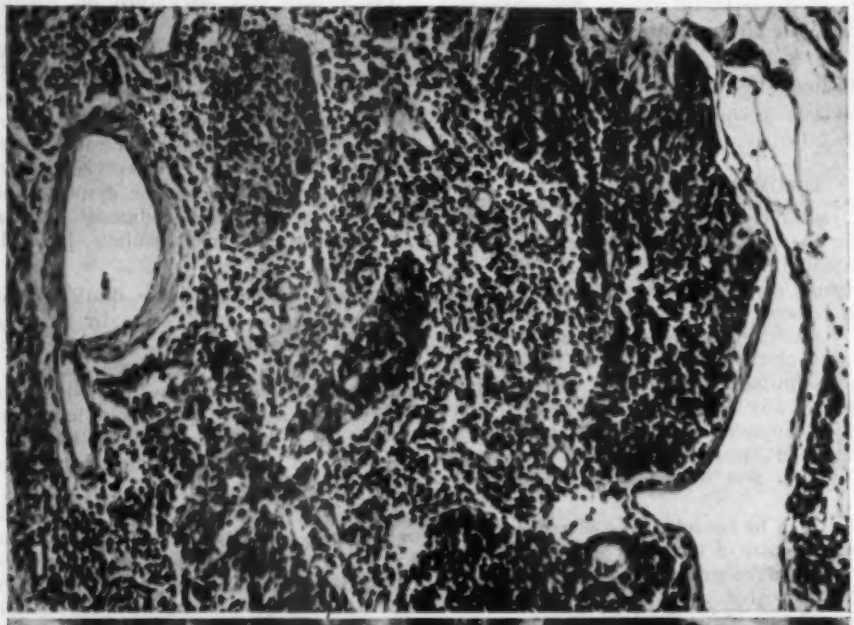
In the left upper part of the mediastinum close to the midline was an encapsulated spherical mass, 2 cm. in diameter, which, on account of its location, consistence and appearance, was diagnosed as hyperplastic goiter with hemorrhage and degeneration. After microscopic examination the diagnosis was changed to neurofibroma.

Within the right obturator foramen a tumor was found, measuring 2 cm. in diameter and weighing 35 Gm., which on the surface and on cut section corresponded completely in consistence and microscopic appearance to the tumor just described.

Further essential observations include: a tumor of the liver, 3 cm. in diameter diagnosed as hemangioma; a tumor of the lower third of the esophagus, 1.5 cm. in diameter, diagnosed as fibromyoma; nodular hyperplasia of the prostate gland; passive congestion of the lungs; arteriolar nephrosclerosis. The skeletal muscles showed no gross abnormalities. The ocular muscles were not examined. The brain showed no gross changes.

Microscopic Description.—The tumor of the thymus was divided into irregular lobules by strands of a dense fibrous tissue. There was no differentiation between the cortex and the medulla. The epithelial reticulum cells and the lymphocytes were distinct (fig. 1). Their distribution and relation to each other varied. There were small and large irregular islands which consisted solely of an epithelial syncytium (fig. 2). The nuclei were round or oval and had a dense chromatin network with one or several nucleoli. The cytoplasm with hematoxylin-eosin stain was pale pink and formed numerous processes which anastomosed with one another. In several small islands the nuclei showed a radial arrangement and gave an acinus-like impression, but no lumen could be detected. In other nests the periphery was moderately infiltrated by lymphocytes. Here the reticular syncytium was not so closely packed and showed the beginning formation of a network, the meshes of which were filled with lymphocytes. In some areas the lymphocytes dominated the picture, being pres-

ent in large collections. But even in those places the epithelial reticulum was recognizable as a meshwork containing lymphocytes. In places the reticulum cells reached a huge size. The nuclei were lightly stained, vesicular, and showed a fine network of chromatin with one or two small distinct nucleoli. The lymphocytes belonged to the small or medium-sized type.



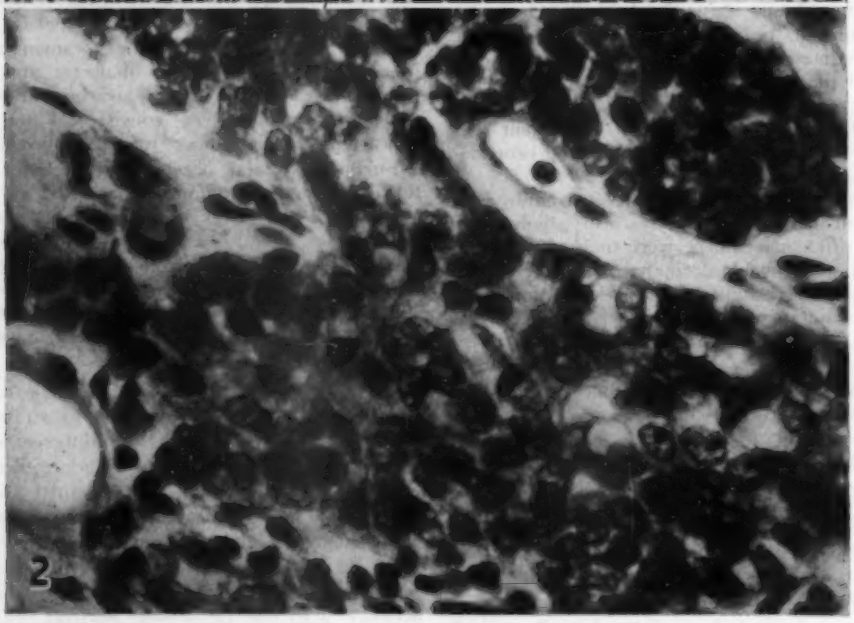


Fig. 1.—Thymic tumor, low power photomicrograph; hematoxylin-eosin stain. Note loss of normal thymic structure. There is fairly regular distribution of epithelial islands in the lymphoid tissue.

Fig. 2.—Thymic tumor; high power photomicrograph; hematoxylin-eosin stain. An island of epithelial syncytium is seen, with only few lymphocytes.

Scattered throughout were eosinophilic granulocytes. Within the fibrous septums and the parenchyma was a moderate number of blood vessels. Sections through the cystic areas showed large empty spaces, and scattered throughout were small accumulations of lymphocytes and reticulum cells. No necrotic material and no distinct wall or capsule could be made out. Sections failed to reveal Hassall's corpuscles, and normal thymic tissue was not recognizable. A small amount of fat tissue was present between and within the lobules. The tumor was considered to be thymoma of a simple type ("considerable thymus hyperplasia," Norris [1]).

Sections taken from the psoas muscle showed a few accumulations of small and medium-sized lymphocytes and plasma cells in the interstices. These collections of cells bore no relationship to blood vessels and were

1. Norris, E. H.: Am. J. Cancer **27**:421, 1936.

thought to be the so-called lymphorrhages. The surrounding muscle fibers appeared normal, and no atrophy or degenerative changes were found.

The myocardium showed moderate brown atrophy with areas of compensatory hypertrophy. In a few areas small "lymphorrhages" were seen.

The tumor from the upper mediastinum showed large areas of necrosis. Where the structure was still recognizable the tumor consisted of wavy and interlacing strands of a fibrillary connective tissue. The nuclei were elongated and arranged parallel to each other, suggesting palisading. Numerous dilated lymph and blood vessels were seen. Many focal areas of recent and old hemorrhage with beginning or advanced organization were scattered throughout the tumor. With Mallory's phosphotungstic acid stain, numerous bluish-stained neurofibrils could be demonstrated, particularly in those areas where the nuclei suggested palisading. Within the fibrous capsule, portions of a peripheral nerve were seen.

The tumor from the obturator foramen was almost identical in structure with the tumor from the upper mediastinum. In view of the general structure, the presence of fibroglia and their close relationship to peripheral nerves, these tumors were diagnosed as neurofibroma.

On the surface of the skin of the chest and the back several "wartlike" raised plaques were seen. Unfortunately, none of these were taken for microscopic examination. In view of the presence of neurofibroma at two sites, these were possibly other manifestations of neurofibromatosis of Recklinghausen.

The cerebral meninges were thicker than average and showed a few scattered perivascular accumulations of round cells and monocytes. The cortex, the white matter and the brain stem showed no abnormalities of structure.

The adrenals showed no appreciable changes.

In reviewing the literature we did not find any other report of neurofibromatosis in a patient with myasthenia gravis. According to Ewing,[2] neurofibromatosis is frequently found in patients with abnormalities of the central nervous system or of the endocrine glands. A multiplicity of tumors in association with myasthenia gravis is mentioned only once by Miller,[3] who reported a case of myasthenia gravis with persistence of a large thymus, hemangioma of the liver and lymphangioma of the spleen.

SUMMARY

In a typical case of myasthenia gravis the autopsy revealed thymoma (marked hyperplasia of the thymus) and neurofibromatosis.

2. Ewing, J.: Neoplastic Diseases, ed. 4, Philadelphia, W. B. Saunders Company, 1940.
3. Miller, H. G.: Arch. Path. 29:212, 1940.

Laboratory Methods and Technical Notes

MYELIN STAINING BY A FIXED SCHEDULE FOR THE OCCASIONAL USER

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For many years I had used Weil's [1] modification of the Weigert myelin method because of its ready applicability to paraffin sections from blocks prepared for the routine methods. Since much of the material requires prompt diagnosis, the value of such a method is apparent.

Usually fairly successful preparations were obtained, but, owing to lack of continued practice, it was difficult for the technician to make regularly myelin stains showing optimum differentiation. The correct point in the differentiation in the iron alum after the hematoxylin stain is difficult for the casual stainer of myelin to discern. Preparations are perhaps more commonly under than over differentiated, and less often just right.

Recently I was faced with the need for a considerable number of myelin preparations in current studies on poliomyelitis. Dissatisfied with the lack of uniformity attainable by our usual method, I initiated the following series of experiments, which resulted in a usable myelin staining method combined with a quite satisfactory cell stain. Further, some interesting basic data were accumulated.

Weil mordanted paraffin sections of formaldehyde-fixed material for ten minutes in 5 per cent potassium dichromate ($K_2Cr_2O_7$). With previously chromated material this step was omitted. Sections were then washed in water and stained for fifteen minutes at 45 to 50 C. in equal parts of 4 per cent aqueous solution of iron alum (ferric ammonium sulfate; $NH_4Fe(SO_4)_2 \cdot 12H_2O$ and aged 1 per cent alcoholic solution of hematoxylin. He then rinsed and differentiated the material in 4 per cent iron alum solution until gray and white substance could be discerned. Then followed another rinse and completion of differentiation in a solution of 1 Gm. borax (sodium tetraborate decahydrate; $Na_2B_4O_7 \cdot 10H_2O$) and 1.25 Gm. potassium ferricyanide ($K_3Fe(CN)_6$) in 100 cc. water to blue-black myelin. Such mixtures will hereafter be referred to as borax-ferricyanide, with the quantities, when necessary, thus: 1:1.25:100.

After a number of unsatisfactory early trials in which we relied on visual differentiation, we decided to fix time, temperature and the concentrations of solutions for all steps in the process except the one then under study. These preliminary trials had indicated that a 1:1,000 solution of safranin O (NS-11)[2] in 1 per cent aqueous acetic acid solution (to be referred to later in this paper as acetic safranin) was a satisfactory counterstain. Results regarding interplay of staining time and iron alum and borax-ferricyanide differentiations had been confusing. The alcoholic solution of hematoxylin had to be aged for some hours but not for any prolonged period.

In the first of these "fixed condition" experiments the material was monkey cerebellum fixed in Orth's fluid. Staining was set at thirty minutes at 55 C. in equal parts of 4 per cent iron alum and one to five day old 1 per cent alcoholic hematoxylin (FH-16).[2] This mixture is always freshly prepared and is referred to hereafter as iron alum–hematoxylin. After staining, sections were washed in running water. Then they were decolorized for one hour in 4, 2, 1, 0.5, 0.2 or 0.1 per cent iron alum. Next they were rinsed, and one set was counterstained at once; the other was treated for eight minutes in Weil's borax-ferricyanide (1:1.25:100) and was then rinsed and counterstained in acetic safranin as previously described. All sections except when otherwise specifically noted were routinely rinsed in water after the counterstain, and dehydrated, cleared and mounted by a sequence of acetone, acetone plus xylene, xylene, xylene and 60 per cent xylene clarite.

Without the borax-ferricyanide step in the preparation, all sections were paler and muddier, with more gray in the background, than the corresponding ones which had been treated with the borax-ferricyanide solution. The latter sections showed progressively deeper myelin staining with decreasing strength of the iron alum. At 4 and 2 per cent iron alum myelin was pale; at 0.2 and 0.1 per cent cells and molecular substance were gray; at 1 and 0.5 per cent contrasts were excellent.

Repetition of this test after the best variations of the other steps had been worked out gave essentially similar results. The staining in iron alum–hematoxylin was carried on for forty minutes at 55 C.; the treatment in borax-ferricyanide (1:2.5:100) was carried on for ten minutes, and the counterstaining in acetic safranin was done for five minutes. The material was formaldehyde-fixed guinea pig brains mordanted two days in 2.5 per cent potassium dichromate and embedded in paraffin. The differentiation was done in 4, 2, 1 and 0.5 per cent iron alum for ten, fifteen, twenty, thirty, forty-five and sixty minutes.

From the Pathology Laboratory of the National Institute of Health.

Technical assistance for this investigation was financed by a "conditional gift" of the National Foundation for Infantile Paralysis.

1. Weil, A.: Arch. Neurol. & Psychiat. **20**:392, 1928.

2. The dye samples used are indicated by the certification lot number given by the Biological Stain Commission or by the manufacturer's initials and lot number. N.A. indicates National Aniline Division, Allied Chemical and Dye Corporation; H. L., Hartmann-Leddon Company; C. B., Coleman and Bell; Biosol, Biosol Products Company; dP, E. I. du Pont de Nemours Company; Gr., Dr. G. Grübler.

Excellent fine and coarse myelin staining with little or no brown or gray in cells was attained at forty-five and sixty minutes in 0.5 per cent and at thirty minutes in 1 per cent iron alum. With stronger iron alum it was difficult to find a decolorizing period which adequately decolorized the background without loss of myelin staining. The prolonged differentiation in weaker iron alum gave the widest margin of safety.

In the second experiment the iron alum–hematoxylin staining at 55 C. lasted ten, twenty, thirty, forty, sixty, ninety and one hundred and twenty minutes. Differentiation was one hour in 1 per cent iron alum, then eight minutes in borax-ferricyanide (1 : 1.25 : 100) and five minutes in acetic safranin.

With ten to twenty minutes of staining, fine fibers were pale and inadequately stained; with sixty to ninety minutes nerve cells were gray and myelin black; at one hundred and twenty minutes everything was deep gray to black. Staining for thirty to forty minutes gave adequate myelin staining and good differentiation.

Next, paraffin sections from monkey brains fixed in Orth's fluid and in dilute aqueous solution of formaldehyde were compared. The latter were brought to water and mordanted at 55 C. in 5 per cent potassium dichromate solution for fifteen and thirty minutes, one, two, four, eight, sixteen and twenty-four hours. Unchromated controls from the brains fixed as described were also used. All sections were then washed ten minutes in running water, stained forty minutes at 55 C. in iron alum-hematoxylin, differentiated for one hour in iron alum (one set in 1 per cent and one in 0.5 per cent iron alum), treated eight minutes in borax-ferricyanide solution (1 : 1.25 : 100) and counterstained in acetic safranin.

Coarse myelin bundles were blue-black and fine fibers were unstained in the unchromated formaldehyde-fixed control and with fifteen minutes to four hours mordanting, while the control fixed in Orth's fluid gave excellent blue-black fine fibers as well. The safranin counterstain was distinctly pale after eight hours' mordanting and disappeared with sixteen and twenty-four hours'. Coarse myelin bundles were paler blue with sixteen hours and unstained with twenty-four hours' mordanting.

An attempt to substitute 1 per cent alcoholic brazilin for the hematoxylin yielded unsatisfactory pale brown myelin. However, the sample was an old Grübler brazilin, and it is possible that fresh dye might be successful.

In the next experiment monkey cerebellums fixed in dilute aqueous solutions of formaldehyde and in Orth's fluid were used; the staining in iron alum-hematoxylin was carried on for forty minutes at 55 C.; differentiation in 0.5 per cent iron alum was done one hour and after treatment in variable borax-ferricyanide the usual five minute counterstain in acetic safranin followed. The variations were these: borax: potassium ferricyanide: water 2:2.5: 100 for two, four and eight minutes, 1 : 1.25 : 100 for two, four and eight minutes and 1 : 0.625 : 100, 1 : 2.5 : 100, 1 : 3.75 : 100, 2 : 0.625 : 100, 2 : 1.25 : 100 and 2 : 1.875 : 100 for eight minutes.

The shorter times were inadequate. One per cent borax was adequate. Two and five-tenths per cent potassium ferricyanide gave better differentiation than lower levels and as good as higher.

In a further test on guinea pig brains borax-ferricyanide 1 : 2.5 : 100 was used for two and a half, five, seven and a half, ten, fifteen, twenty, twenty-five and thirty minutes. All other steps were the same as in the last experiment.

At two and a half minutes cells were gray; with longer times, pink. Myelin was grossly blue-black at two and a half to ten minutes and still well stained microscopically up to thirty minutes. Material fixed primarily with formaldehyde and postchromated after blocking gave quite uniform myelin staining throughout, while that fixed in Orth's fluid gave better stained myelin near the surfaces of the brain.

For the next experiment the brain of a monkey was fixed whole within thirty minutes after death in 1 part 37 per cent formaldehyde solution plus 9 parts 1 per cent sodium chloride solution. This was changed on the thirteenth day to buffered (p_H 7.0) 3.7 per cent formaldehyde solution, and on the twenty-fourth day a series of fifteen blocks of cerebellum were cut. One was embedded in paraffin without chromation, one was mordanted in 2.5 per cent potassium dichromate for two days (a routine procedure), five were mordanted in 5 per cent potassium dichromate for one, two, four, seven and fifteen days respectively, 8 were mordanted in Weigert's first mordant (potassium dichromate 5 per cent, chromium fluorate [CrF_3] 2.5 per cent), two sets for two, four, seven and fifteen days; one of these sets was then treated two days more in Weigert's second mordant (chromium fluorate 2.5 per cent, copper acetate 5 per cent, glacial acetic acid 5 per cent, water 100 per cent). Paraffin sections of all were stained forty minutes at 55 C. in iron alum-hematoxylin, differentiated one hour in 1 per cent iron alum and ten minutes in borax-ferricyanide 1 : 2.5 : 100 and counterstained five minutes in acetic safranin.

Nuclear staining was excellent in material mordanted two days in 2.5 per cent or one to seven days in 5 per cent potassium dichromate; it was impaired with fifteen days. In comparison nuclear staining in the material mordanted two days in Weigert's first mordant was inferior and became worse with longer exposure, either with or without Weigert's second mordant.

Myelin staining was best in the block mordanted four days in 5 per cent potassium dichromate and in that treated two days each in Weigert's first and second mordants. Mordanting two, seven and fifteen days in 5 per cent potassium dichromate or four and seven days in Weigert's first mordant followed by two days in his second gave nearly as good results. Next came the block treated two days in 2.5 per cent potassium dichromate, that given one day in 5 per cent potassium dichromate and that given four days in Weigert's first mordant alone. Inferior myelin staining was obtained in blocks mordanted two and seven days in Weigert's first mordant, and the poorest followed fifteen days in Weigert's first mordant, with or without his second.

It appears that optimum cellular detail combined with the best myelin staining was attained in this series by mordanting two to four days in 5 per cent potassium dichromate.

Attempts to substitute buffer solutions for the iron alum differentiator gave inferior results. Buffers between p_H 2.9 and 5.0 largely decolorized the myelin; those between 6, 4 and 7.6 left too much gray in cells and background. At p_H 5.5 myelin was a little pale; at 6.0 cells were slightly gray as compared with the iron alum control.

In these counterstaining experiments forty minutes at 55 C. in iron alum–hematoxylin, one hour in 0.5 per cent iron alum and eight minutes in borax-ferricyanide 1 : 1.25 : 100 were tried. Picro aniline blue (LK-1), picro fast green FCF (NGf-3), picro wool green S (dP1940) and picro acid fuchsin were tried at 100 mg. per hundred cubic centimeters of saturated aqueous

picric acid. These mixtures were used alone for five minutes. The first three were used also with a preceding acetic safranin stain for two minutes, staining for three minutes. These same three were also employed with preceding plasma stains for four minutes, 0.2 per cent Biebrich scarlet (HL, 1939) or 1 per cent orange G (Gr. 12.37) in 1 per cent acetic acid, followed by a 1 per cent acetic acid rinse and four minutes in the picric acid mixture. All the picric acid stains were differentiated with 95 per cent alcohol.

The safranin nuclear stains were not evident in the second group. In the first and second groups the blue, green or red dye dominated, as also in the orange G plasma stain group. The Biebrich scarlet dominated over or blended with the blue or green connective tissue stains. Apparently, connective tissue stains of the picric acid type do not combine well with myelin stains.

Substitution of an azure A eosinate stain at p_H 4.0, 4.6, 5.6 and 6.5 for the acetic safranin yielded quite acceptable results at p_H 4.0 and 4.6. Contrasts were not as good as with acetic safranin.

In another series the same iron alum–hematoxylin stain and one hour 0.5 per cent iron alum differentiation were used with a ten minute borax-ferricyanide step at 1:2.5:100 and followed by five minute stains in 1:1,000 solutions in 1 per cent acetic acid of a series of basic aniline dyes. The material was formaldehyde-fixed guinea pig brains chromated two days in 2.5 per cent potassium dichromate. Sharp blue chromatin and tigroid with fair contrast to the greenish black myelin were found after use of new methylene blue (NA no. 3217), thionin (NT-8), toluidine blue (NU-2 and NU-3), azure C (NAc-2) and azure A (NAz-8). Greener blue chromatin and paler tigroid were observed with azure B (NA no. 7724) and methylene blue (NA-24). Crystal violet (LC-12), malachite green (NMg-7), victoria blue B (Biosol: C.I. no. 729), new fuchsin (NA no. 8963), rosaniline chloride (NA no. 9543), basic fuchsin (NF-28) and pararosaniline acetate (NA no. 8772) all overstained cells and ground substance and tinged myelin, the last least. Acceptable nuclear stains were obtained with nile blue A (Grübler?), methyl green (CG-8), janus green B (HL, 1930), and bismarck brown Y (NN-6), but the best stains were methylene violet (C. I. no 842, Hoechst), safranin 6 B (C. I. no. 843; NA no. 10071) and safranin O (NS-11). These three gave, respectively, bluish red, red and orange-red chromatin and tigroid. The last afforded the most brilliant contrast with the blue-black myelin. Seven other samples of safranin O gave similar results.

The foregoing tests were done on monkey and guinea pig brains fixed in Orth's fluid or in dilute aqueous formaldehyde solution with and without postchromation. When this technic was applied to a human brain (case 15) only fair to poor myelin staining was obtained, though the differentiating fluid was decreased to 0.25 per cent, so that red corpuscles and cerebellar granule cells stained black. This material had been chromated as usual, two days in 2.5 per cent potassium dichromate. Prior to chromation it had been stored for more than six years in dilute formaldehyde solution. There was evident also moderate postmortem autolysis. As it was considered that the failure of myelin staining might be due either to autolysis or to long storage, another brain (case 2), showing a similar grade of autolysis, was selected. This brain had been in formaldehyde solution only two months. From these two brains series of blocks of cerebellar cortex were cut. One of each was embedded in paraffin without chromation; others were mordanted in 5 per cent potassium dichromate for one, two, three, four, five, six, eight, ten, twelve and fourteen days. All were rinsed one hour in water, soaked sixteen hours in 80 per cent alcohol, dehydrated with acetone, cleared in benzene and embedded in paraffin. Sections were stained forty minutes at 55 C. in iron alum–hematoxylin and decolorized one hour in 2, 1, 0.5 and 0.25 per cent iron alum, also thirty minutes in 0.5 and 0.25 per cent iron alum. All these variants were given next a ten minute treatment in borax-ferricyanide 1:2.5:100, and other slides with the two lower iron alum concentrations received a five minute bath in the aforenamed solution. The usual five minute acetic safranin counterstaining followed.

Excellent myelin staining was obtained in the recent material (case 2) by using 0.5 per cent iron alum for fifteen to sixty minutes and treating for ten minutes in borax-ferricyanide. Uniformly poor staining was obtained in the six year old material, regardless of the variations in staining technic or in the length of chromation. Chromation over eight days reduced the resistance to decolorization of the myelin in the recent case, so that sections treated one hour with 0.5 per cent iron alum were too pale.

Effect of Prolonged Storage in Formaldehyde Solution on Staining of Myelin by a Standardized Technic

		Conditions Giving Best Myelin Stain				
		Iron Alum		Sodium Borate Potassium Ferricyanide, Minutes	Grade of Best Myelin Stain Obtained	
Case	Storage Time	Per Cent	Minutes			
1	13 days	0.5	60	10	Excellent	A
2	2 mo.	0.5	60	10	Excellent	A
3	2-3 mo.	1	60	10	Excellent	A
4	2-3 mo.	0.5-1	60	10	Excellent	A
5	6½ mo.	0.5	60	10	Excellent	A
6	6½ mo.	0.5-1	60	10	Excellent	A—
7	10¾ mo.	1	60	10	Good	B+
8	1 yr.	0.5	60	5	Good	B
9	1½ yr.	0.25	60	10	Good	B
10	2⅛ yr.	0.5	30	10	Fairly good	C+
11	2⅞ yr.	0.5	30	10	Fair	C
12	3 yr.	0.5	60	5	Good, irregular	B—
13	3⁵/₁₂ yr.	0.5	30	5	Fair	C
14	4¾ yr.	0.25	60	5	Poor	D+
15	6¼ yr.	0.25	60	5	Fair	C—

Ten sections from each case were stained for forty minutes at 55 C. in iron alum hematoxylin. Sections were differentiated an hour in 2, 1, 0.5 or 0.25% iron alum, or thirty minutes in 0.5 or 0.25%, and ten minute treatments with 1% $Na_2B_4O_7$ + 2.5% $K_4Fe(CN)_6$ were given (6 sections), or in the 2 lower iron alum levels, also five minute treatments (4 sections). All sections were counterstained five minutes in 1:1000 safranin in 1% aqueous acetic acid solution.

Following this experience, sections of the routine material which had been chromated two days in 2.5 per cent potassium dichromate were selected from these and 13 more cases. From the results in the accompanying table it is evident that storage in formaldehyde solutions for much over six months is distinctly inimical to subsequent staining of myelin. On more than two years of such storage myelin staining is generally poor.

In contrast, two brains originally fixed in Orth's fluid and stored in 80 per cent alcohol for five and a half and eight and a half years respectively still gave quite good myelin staining, though not as good as paraffin sections cut in the week after fixation. Likewise, the sections of the material stored in alcohol were more easily decolorized, requiring thirty to sixty minutes in 0.25 to 0.5 per cent iron alum as compared with the hour in 0.5 to 1 per cent iron alum required by the material stored as paraffin sections.

It is to be noted further that when the optimum time and concenration of the iron alum bath have been determined on one block from a given case or batch of material, the same detailed technic generally yields quite acceptable results on the remaining blocks.

SUMMARY

Acceptable routine staining of myelin may be attained with fixed times, temperatures and solution concentrations by using a variant of Weil's modification of the Weigert myelin stain. Material fixed in dilute aqueous formaldehyde solution and not over six months old should be mordanted two to four days in 5 per cent potassium dichromate solution before dehydration. Chromation of paraffin sections of material fixed in formaldehyde is futile. In this material the coarser myelin bundles can be stained without chromation, and no amount of mordanting restores the fine fibers. Material stored longer than six months in dilute formaldehyde solutions shows a gradual impairment of myelin staining until in three or more years satisfactory stains are not obtainable by any variant tried. Prolonged chromation, whether in the block or on the slide, impairs first staining of chromatin and tigroid and then that of myelin as well. Weigert's potassium dichromate–chromium fluoride is more harmful in this respect than 5 per cent potassium dichromate.

The following staining schedule is suggested.

1. Stain forty minutes at 55 C. in iron alum–hematoxylin (equal parts of one to five day old 1 per cent alcoholic hematoxylin and 4 per cent iron alum). Wash in water.

2. Differentiate one hour in 0.5 per cent iron alum. Wash in water.

3. Treat ten minutes in an aqueous solution containing sodium borate, 1 per cent, and potassium ferricyanide, 2.5 per cent. Wash in water.

4. Counterstain five minutes in 1:1,000 safranin O in 1 per cent acetic acid. Wash in water.

5. Dehydrate, clear and mount, using a sequence of acetone, acetone plus xylene, xylene two changes and 60 per cent xylene clarite.

Methylene violet (C. I. no. 842), various thiazine dyes, janus green B or bismarck brown Y may be substituted as counterstains, but safranin O gave the best contrast.

Historical Review

ORIGINS OF THE CELL CONCEPT IN PATHOLOGY

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A popular magazine recently listed a hundred books considered by philosophers, scientists and educators to be the greatest ever written. In that assembly of masterpieces, which included Copernicus' "De Revolutionibus Orbium Coelestium," Harvey's "De Motu Cordis," Newton's "Principia" and Darwin's "Origin of Species," was placed Virchow's "Die Cellularpathologie." [1] And this because present concepts of disease are still founded on the principles therein set forth. An awareness of this is most keen in the pathologist who daily weighs the significance of cell structure as it reflects the extent and the ultimate outcome of disease.

Like the cells he described, Virchow's book did not arise de novo. It represents the culmination of a long series of researches carried out not only by the celebrated author but by many whose names have been forgotten. The modern scientist and physician, ever on the lookout for the new and the rare, is interested only in the concepts which he still happens to regard as true and not in the manner in which they developed. "We regard the hypotheses of the time as so many truths, and condemn only the theories of the past." This ignorance of the history of one's basic beliefs makes them mere dogmas and places one on an intellectual level with the little boy who, standing by the shore of the sea, thinks it to be everywhere but 2 feet deep.

In this paper an attempt will be made to trace the evolution of the ideas expressed by Virchow in 1858. It will first be necessary to examine the instruments and technics available to investigators during the first half of the nineteenth century. Only by realizing the limitations of their methods is it possible to perceive the source of their errors and to evaluate the magnitude of their achievements.

THE MICROSCOPE

By 1838 the microscope had been changed from the mediocre instrument that it was in 1800 to an instrument of great precision and excellent optical qualities. Although Robert Hooke used a compound microscope, most other seventeenth century workers—e. g., Leeuwenhoek and Swammerdam—used a single lens. During the eighteenth century and the early decades of the nineteenth century the usefulness of the compound instrument was still so impaired by its marked spherical and chromatic aberration that it was almost wholly neglected. In a paper on the red blood corpuscles published in 1777 William Hewson [2] wrote:

> It is by the microscope alone that we can discover these particles; and as some dexterity and practice are required in the use of that instrument, there have not been wanting men of character and ingenuity who, having been unsuccessful in their own experiments, have questioned the validity of those made more fortunately by others. Some have gone so far as to assert that no credit can be given to microscopes; that they deceive us by representing objects different from what they really are.
>
> These assertions, though not entirely without foundation when we speak of one sort of microscope, are very unjustly applied to them all. In compound microscopes when the object is viewed through two or more glasses, if these glasses be not well adapted to the focus of each other, the figure of the object may be distorted; but no such circumstance takes place when we view an object through a single lens. All who use spectacles agree that the figures of objects appear the same to them, as they do to the naked eye. And as the single microscope has, like the spectacles, but one lens between the eye and the object, there is no reason to suppose that one can deceive us more than the other. The compound having a larger field, is more pleasant than the single microscope for many purposes; but the single should be always preferred by those who wish to ascertain the figures of minute bodies. . . .

Though achromatic objectives were made of flint and crown glass in 1733 by Chester Hall, it was not until 1807 that Herman van Deyl produced a satisfactory compound microscope with an achromatic objective and a magnification of 229 ×. Because of the difficulty of constructing achromatic lenses with a short focal distance, much increase in magnification was not achieved. Thus the objective of van Deyl had a focal distance of 13 mm. and an initial magnification of

1. Virchow, R.: Die Cellularpathologie, Berlin, A. Hirschwald, 1858; ed. 2, 1859, (*a*) p. 360; (*b*) p. 359.

2. Hewson, W.: An Experimental Inquiry into the Properties of the Blood: Containing a Description of the Red Particles of the Blood, London, T. Longman, 1777; reprinted in The Works of William Hewson, F. R. S. edited by G. Gulliver, London, Sydenham Society, 1846.

19 ×, compared with the uncorrected lens which had a focal distance of 2 to 3 mm. and an initial magnification of 80 to 100 ×. The final power of the achromatic microscope was therefore gained mainly by means of the ocular, resulting in empty magnification. The advantage lay in the greater numerical aperture of the lens and hence increased brilliance of the image.

In 1825 Chevalier placed Canada balsam between the crown and the flint glass, preventing refraction of the light rays at the line of junction. The focal distance was 37 mm., but the advantage lay in that several of these lenses could be placed together, forming a single objective of higher power and diminished spherical and chromatic aberration. The convex surface of the compound lens was still placed downward, permitting considerable distortion of the image. Amici, working in Italy, produced an objective in 1827 consisting of three crown-flint lenses screwed one on top of the other with the flat surface down. The plane surface of the plano-convex ocular was turned upward. This position of the lenses resulted in the removal of most of the spherical as well as of the chromatic aberration, producing an aplanatic as well as an achromatic lens system. Harting's [3] book on the microscope, published in 1859, is authoritative for this phase of the development of the instrument.

By 1830 the compound microscope as a scientifically accurate instrument was available to a few investigators. In less than a decade, plants for its large scale manufacture were established in England, Germany, France and Italy, and a little later in the United States (Spencer). The eager student, beginning his career in those days when "fundamental discoveries about animal tissues could be made on scrapings obtained by using the edge of a scalpel or even a fingernail," was in a position to purchase the wonderful instrument that made all this possible.

Before leaving the subject it is well to point out that the microscope of 1838 to 1858 was still far from the perfected instrument of today. This is apparent from the words of Flemming [4] written as late as 1882:

> . . . Until a few years ago everyone who worked with powerful lenses—even good ones, such as the higher water immersion systems of Hartnack or Zeiss—probably complained as I did that in spite of the excellent resolution he had to work in twilight. It was an axiom: the stronger the system, the less light. . . . In the meantime an outstanding improvement in microscopic apparatus had been made. This lies in the reintroduction of the homogeneous immersion system by Zeiss, Seibert and Andri, and in the regulated illumination of Abbé. . . . Whoever looks about in the history of histology realizes that important discoveries and theoretic advances were usually not the forerunners of improvements in instruments and technique, but rather their result. . . . It will be due in the first place to the opticians if from the present modest beginnings a real morphology and biology of the cell is developed.

3. Harting, P.: Das Mikroskop: Theorie Gebrauch, Geschichte und gegenwärtiger Zustand desselben, translated by F. W. Theile, Braunschweig, F. Vieweg u. Sohn, 1859.

4. Flemming, W.: Zellsubstanz, Kern und Zelltheilung, Leipzig, F. C. W. Vogel, 1882.

HISTOLOGIC TECHNIC

The modern pathologist, examining only thin, transparent, multicolored sections of tissues, may find it difficult to comprehend why some of the discoveries which now appear so obvious were not made earlier. It is essential for an understanding of the views expressed by the investigators of the period ending in 1858 to realize that a histologic technic as pathologists understand it today did not exist. Bits of tissue were cut off with scissors, then teased out in a drop of water, saline solution or sugar solution and examined.

In 1842 Schleiden [5] wrote that there existed no book on microscopic technic. He suggested that a heavy razor be used to cut sections, the tissue being held between thumb and forefinger. If it were thin—e. g., a hair, a leaf or moss—it could be fastened to the thumb nail with saliva and then sectioned by rocking the blade over the nail, carrying it slightly backward each time. For soft plant and animal tissues he advised embedding in a gum, which was allowed to dry almost to glassy hardness. Thin shavings were then scraped off and placed in water where they were thought to regain most of their former appearance.

Microtomes for cutting thin sections were first introduced for the study of wood about 1750. Some improvements were made in this instrument during the following century, but it did not become popular. Men like von Mohl expressed disdain for investigators who could not cut free hand sections. This was of course partly due to the fact that without a suitable embedding technic thin sections of soft tissues could not be cut even with the aid of a microtome. The rotary instrument, now almost universally used, is an American invention. It was introduced independently in 1885 by Pfeifer of the Johns Hopkins University and Minot of Harvard.

Most early embedding matrices only served to enclose the tissue; they could not be made to permeate it. This was accomplished in 1873

5. Schleiden, M. J.: Grundzüge der wissenschaftlichen Botanik nebst einer methodologischen Einleitung, Leipzig, W. Engelmann, 1842.

by Flemming, who introduced soap for the purpose. Though paraffin was used in 1876, it was not entirely satisfactory until chloroform replaced turpentine as the solvent in 1881. Duval employed celloidin as an embedding matrix in 1879.

Fixing fluids were at first used to achieve hardening of the tissues and facilitate sectioning, rather than to preserve the cells. One of the first used was chromic acid. Though acetic acid fixed the tissues, it was valued chiefly because it made the nucleus visible in fresh preparations. Most common fixing solutions were introduced during the decade from 1880 to 1890.

When the pathologist now thinks of a histologic preparation his mind's eye presents him with a gaudy picture in which red and blue are the dominant colors. Yet, though Hill had used cochineal in the study of plant anatomy as early as 1770, and Corti had employed carmine casually in his study of the cochlea in 1851, Gerlach began the modern tinctorial era in 1858. Many are familiar with the happy accident which brought this about. Through an oversight Gerlach had left a section of cerebellum, hardened in potassium bichromate, in a very dilute solution of carmine overnight. When he found the section the next day, it was beautifully stained and showed excellent differentiation of nerve fibers and cells. In 1865 Boehmer brought forward hematoxylin in its present form; ten years later Fischer used eosin as a counterstain. An excellent bibliography of the subjects here touched on is given in a paper by G. M. Smith.[6]

To gain some insight into the appearance of tissues when examined after using the technics employed by the early histologists, the reader might well cut a few extra sections the next time he uses the freezing microtome. Without clearing or staining, the sections may be mounted in saline solution and inspected. All the discoveries that I shall detail were made on preparations that were even thicker and more distorted; the very cover glass has since been perfected!

THE CELL THEORY IN 1838

Before considering the development of the cell concept in pathology, it is necessary to review briefly the state of things in 1838 and how it had come about. In recent years several writers have been greatly exercised over the attention given the work of Schleiden and Schwann, to the neglect of others, notably Dutrochet.

Speculation on the structural elements of animal tissues is as old as philosophy. Aristotle had a hand in this as in nearly everything else; and an allusion to the "atomic" nature of the body is found in Lucretius' poem "De Rerum Natura." About the middle of the eighteenth century Albrecht Haller supported and elaborated the theory that all animals and plants are made of fibers, the interspaces filled by a kind of glue. This theory received widespread acceptance by reason of the authority of the man who sponsored it. Not long afterward, however, many investigators began to insist that the elementary bodies were granules or globules instead of fibers, basing their belief on direct observation.

But microscopic examination of nearly any tissue, using direct sunlight for illumination as was then the custom, will lend it a granular character due to the refraction and dispersion of the light rays. In 1826 Milne-Edwards[7] wrote of the fibers:

> . . . Using a more powerful instrument I found that these cylinders . . . are themselves made up of spherical corpuscles arranged in an irregular row and having a diameter of 1/300 mm.

Another Frenchman, Raspail,[8] writing at that time, was likewise a believer in the existence of elementary bodies. These he thought arose in the form of oil droplets which assumed a spherical shape in water and on contact with atmospheric air took up carbon dioxide. He compared these cells with crystals and called their mode of formation a crystallization.

Meanwhile, botanists had made great strides toward a true understanding of the structure of plants. Since the days of Leeuwenhoek, Nehemiah Grew and Robert Hooke they had been conscientiously drawing cells in all their figures of the cross sections of stems, roots and leaves. But Charles Francois Mirbel[9] was among the first to insist that most if not all parts of a plant are made up of cells, and his illustrations go far to prove the point. In 1802 he stated that cells make up the whole of all mushrooms and the cortex of monocotyledons and dicotyledons, as well as the pith of all plants. The parenchyma of flowers and buds, of stamens, pistils and pollen consists of pigmented cells. Of the mode of their development he said only, "The cells and tubes appear as a result of the imbibition of water, but the details are unknown."

Another botanist, L. C. Treviranus, was likewise a champion of the importance of the cell in plant structure and demonstrated the cellular nature of the spiral vessels. Among the greatest of these plant histologists was Hugo von Mohl,

6. Smith, G. M.: Tr. Am. Micr. Soc. **34**:71, 1915.

7. Milne-Edwards, M. H.: Ann. d. sc. nat. **9**:362, 1826.

8. Raspail, F. V.: A New System of Organic Chemistry, translated from the French by W. Henderson, London, 1834.

9. Mirbel, C. F.: Traite d'anatomie et de physiologie végétales, Paris, 1802.

who as early as 1837 demonstrated the development of new cells in algae by the formation of cross walls.[10]

Of the utmost importance for the subsequent development of the cell concept was the discovery of the nucleus. This was announced in a paper delivered before the Linnean Society of London in 1831 by Robert Brown.[11] The structure had been noted before; e. g., the Italian Fontana[12] described nucleated cells in the slime scraped from the skin of eels. Other botanists had seen it too; but, as Brown himself said, "So little importance seems to be attached to it that the appearance is not always referred to in the figures in which it is represented." Brown noted its presence in the most diverse tissues of a single plant, as well as in many different species of plants.

In 1835 Johannes Müller, professor of pathology, physiology and comparative anatomy at the University of Berlin, published his observations on the anatomy of lampreys.[13] Writing of the structure of the notocord, he said:

> . . . The jelly it contains, when cut in thin slices and examined under the microscope is found to be homogeneous, translucent, not granular, and traversed by a network of cell walls. The cells are dissimilar and irregular, but are like the plant cells in that the walls are everywhere closed and usually abut in straight lines, presenting unequal many-sided figures in cross section. The jelly in the cells is like the walls of the cells themselves.

The significance of this quotation lies in Müller's recognition of the similarity that existed between these structures in an animal and those previously observed in plants. Schwann was then one of his pupils, and it was at the suggestion of Müller that he started work on the cellular character of animal tissues, beginning with a study of the blood vessels.

The activity in the field of animal microscopy at that time is indicated by the number of important contributions made in 1836. Valentin found nucleated spherules in the abdominal nerve strand of the leech which were similar to those observed in the ganglions of vertebrates. Related structures were described in the ganglions of other invertebrates by Ehrenberg. Purkinje saw "tailed corpuscles in the yellow mass between cortex and medullary substance of the cerebellum." Henle had previously shown that the cilia of oysters were attached to the cylindric nucleated bodies that formed a single layer, standing one beside the other. On further investigation he found nucleated cells not only in all ciliated mucous membranes of higher animals but in the epithelium of the intestinal tract as well. Formerly the epithelium had been thought of as a secretion on free tissue surfaces; this work of Henle's helped to overthrow that idea.

In the following year a young Frenchman, P. J. F. Turpin, made similar observations on the epithelium of the vagina.[14] He concluded:

> . . . One is not able to deny, after having studied well the vesicles of which this layer of mucus formed by the vaginal mucous membrane is composed, that they form a well organized cellular tissue consisting like the cellular tissue of plants of an aggregation by simple contiguity of separate vesicles.

Another Frenchman, Dutrochet, abreast of the advances being made, attempted to formulate a general theory on the ultimate structure of animals and plants based on these investigations. That he came close to the truth is seen in the following paragraphs from his book[15] published in 1837:

> . . . One general fact seems to have emerged from the microscopic observations made upon animal structures; this fact is that all organs are composed of very small globules, often appearing grouped in confusion, at other times arranged in rectilinear series. These globules are generally 1/300 mm. in diameter according to M. Milne-Edwards; but this assertion should not be generalized, for observation shows that these globules vary greatly in size. Some are much larger, others smaller, almost beyond range of the microscope. Finally, they are occasionally no longer even visible, although their existence founded on analogy cannot be in doubt.
>
> The globules that form by their aggregation the majority of the organs of animals are small membranous vesicles. One sees that nature possesses a uniform plan for the structure of animals and plants. In all these organisms the minute structure is a conglomeration of utricles which occasionally are globular, sometimes elongated, or reduced to a simple sphere of extreme smallness. These utricles greatly resemble one another in all animals.

Apparently there is here a completed "cell theory" a year before the papers of Schleiden and Schwann were published. Yet it exerted little influence on contemporary biologists, primarily because it differed little from the generalizations which had preceded it. The work of Schleiden and Schwann was unique in that they stressed the importance of the nucleus, so ably described seven years before by Robert Brown. In all previous discussions the elementary bodies were called granules, globules, vesicles, and had no definite morphologic character-

10. von Mohl, H.: Flora **20**:1, 1837.

11. Brown, R.: Tr. Linnean Soc., London **16**:685, 1833.

12. Fontana, F.: Treatise on the Venom of the Viper, on the American Poisons, and on the Cherry Laurel, and Some Other Vegetable Poisons, translated by J. Skinner, London, J. Murray, 1787.

13. Müller, J.: Vergleichende Anatomie der Myxinoiden, Berlin, 1835.

14. Turpin, P. J. F.: Ann. d. sc. nat. **7**:207, 1837.

15. Dutrochet, R. J.: Mémoires pour servir à l'histoire anatomique et physiologique des végétaux et des animaux, Paris, J.-B. Baillière, 1837, vol. 2.

istics. Thus the work of Dutrochet is marred by the vagueness of his concept of cell structure.

By insisting on the presence of a nucleus Schleiden and Schwann removed at one stroke the whole ambiguous mass of granules and globules which had for so long been the favorite sport of "systematizers" and which were scarcely more specific than the "atoms" of Lucretius. Furthermore, they stressed the importance of the nucleus in cell multiplication, though their concept of the manner in which this occurred was erroneous. The term "cell theory" when applied to the work of Schleiden and Schwann should emphasize their description of cell structure and multiplication rather than the concept of the cell as a structural unit common to plants and animals. The latter was admittedly promulgated by many earlier investigators.

Schleiden [16] thought that within the large cell at the end of the pollen tube he had observed the formation of additional nuclei by the aggregation of granules. Along one side of the young nucleus a blister-like elevation then developed which gradually became larger and finally increased so much in size that the nucleus appeared as a large granule attached to the inner surface of the blister.

This report stimulated Schwann to search for a similar mode of cell multiplication in animals. But he went further than Schleiden and claimed to have seen it not only within preexisting cells but even in free fluids and exudates.[17]

> . . . The following admits of universal application to the formation of cells: there is present at first a structureless substance which is sometimes quite fluid, at others more or less gelatinous. This substance posesses within itself . . . a capacity to occasion the production of cells. When this takes place the nucleus usually appears to be formed first, and then the cell around it. The formation of cells bears the same relation to organic nature that crystallization does to the inorganic. . . . This is the fundamental phenomenon of all animal and plant growth. It is alike equally consistent with those instances in which young cells are formed within parent cells as with those in which formation goes on outside them. The generation of the cells takes place in a fluid or in a structureless substance in both cases. We will name this substance in which the cells are formed, cell-germinating material or cytoblastema.

THE CELL IN PATHOLOGY

It is remarkable and certainly significant that four leaders in the new field of investigation were closely associated with one another at the time of its inception. Henle and Schwann occupied rooms in the same boarding house, and both were students of Johannes Müller at the University of Berlin. Virchow, who was also one of the latter's students, said [18]:

> . . . He partly founded my interest in anatomy, and determined it forever. I enjoyed his teaching, his council, and his fatherly friendship for a year and a half.

Elsewhere [19] he wrote:

> . . . I began my studies in the same year that Schwann, after publishing his work, accepted a call to the Catholic University of Louvain. The scientific atmosphere was still wholly filled with the new thoughts. Johannes Müller himself, our revered master, discussed them fully in his teachings. In fact, he was the first to give the cell concept a broad application to pathology in that he directed it upon the study of tumors. What wonder then that we younger men early learned to think in terms of cellularity.

After several timid and fleeting appearances during the previous two years, the cell made its grand entry into pathology in 1838 with the publication of Müller's "On the Minute Structure and Form of Morbid Tumors." In this book the author refers to the work of Schwann with the words:

> . . . As a result of these investigations I even examined tissues in which no cells had yet been found, for their presence. . . . In several tumors in which until then I had not seen any cells, I now discovered them.

He agreed with the prevailing concept that neoplasms were primarily degenerative processes. To this he added the new idea that with the degeneration of normal tissue there is associated a new formation of elements that replace the healthy structures. The formless blastema of Schwann led Müller to say: [20]

> . . It can be clearly shown that the germinal cells of carcinoma do not arise from preexisting fibers but develop independently from a true morbid fluid between the tissue cells.

He was convinced that tumor cells could arise not only within the cytoplasm of preexisting cells (endogenous cell formation) but also within a structureless exudate (free cell formation). He thought that these findings in tumors confirmed Schwann's views on the genesis of cells. Of the structure of tumors he said further:

> . . . The finer microscopic elements of tumors consist of capillaries, fibers, granules, cells with and without nuclei, tailed and spindle-shaped corpuscles and vessels. . . . By far the most common element of tumors is the cell. This is the case in sarcoma, cellular enchondroma and carcinoma simplex, reticular as well

16. Schleiden, M. J.: Arch. f. Anat. u. Physiol. **5**: 137, 1838.

17. Schwann, T.: Microscopical Researches into the Accordance in the Structure and Growth of Animals and Plants, translated from German by H. Smith, London, Sydenham Society, 1847.

18. Virchow, R.: Johannes Mueller—Eine gedächtnissrede, Berlin, A. Hirschwald, 1858.

19. Virchow, R.: Virchows Arch. f. path. Anat. **87**: 389, 1882.

20. Müller, J.: Ueber den feineren Bau und die Formen der krankhaften Geschwülste, Berlin, G. Reimer, 1838.

as alveolar. It is difficult to recognize them at lower magnifications, for then they look like granules, but when higher powers are used most tumors resolve themselves into cells.

Müller pointed out that some tumors such as the cholesteatoma consist solely of cells but that in others—e. g., many kinds of carcinoma—the cells are present in large masses separated by fibrous bands.

The "tailed corpuscles" of Müller were at once seized on by many microscopists as the cell specific for all malignant tumors. Yet Müller's original description of them should have left no doubt of their real nature:

> . . . Another element frequently found in tumors is the tailed corpuscle which, as I pointed out, occasionally occurs in sarcoma and melanoma. . . . The tailed corpuscles are not peculiar to the sarcoma, for I have repeatedly seen sarcomas in which they are entirely absent. They also occur quite as frequently in tumors that are not cancerous.

Despite these clear statements, the *geschwänste Körperchen* were the subject of heated disputes for a decade. With regard to a specific cancer cell, Müller had at once pointed out that "the carcinoma is not a unique tissue, and its finest elements do not differ markedly from those of benign tumors and the primitive tissue of embryos."

Yet Müller was a bit too pessimistic as to the diagnostic value of his findings when he wrote:

> . . . The diagnosis must be based on easily recognizable characteristics that require no special talent and no skill of a physiologist. Microscopic and chemical analysis will therefore never become a means of medical diagnosis; it would be ridiculous to wish this or even consider it possible.

On Feb. 16, 1838, Jacob Henle[21a] read a paper before a medicosurgical society, entitled "Concerning the Formation of Mucus and Pus and Their Relation to the Epidermis." At that time he was living with Schwann, while the latter's histologic studies were appearing in *Froriep's Notizen*. Henle began with a general discussion of epithelium and its distribution in the body and summarized his observations with these words: "It is a single or stratified layer of cells which covers all surfaces of the body, lines its canals, tubes, and the walls of its cavities." His presentation of the formation of pus was cautious. On the basis of studies carried out on nasal secretions during an attack of coryza, Henle stated that first the epithelial cells are desquamated. Within the fluid secreted on the denuded surface there then appear granules which aggregate to form nuclei. These in turn develop into the so-called primary cells which are changed into pus corpuscles. The latter ultimately give rise to new epithelium.

A year later Henle amplified his views[21b] and cited the work of Julius Vogel, who had apparently observed all these changes in a rabbit's ear which had been treated with cantharidin. Henle concluded that a plastic exudate (blastema) is formed in all healing and regenerative processes. Within this are first developed the pus cells, which, however, are only a primitive and undifferentiated stage in the development of more complex tissues, such as bone, nerve or epithelium. The primitive corpuscles are produced in excess and are washed away as "pus."

How completely the schwannian concept of free cell formation in a blastema was accepted by pathologists is shown in Henle's discussion of the different kinds of exudate. Depending on the further development of the primary cells and on the quantity and the quality of the exudate, the subsequent course of inflammation was one of three types.

1. The quantity of the exudate is small. In this develop primary cells that are directly converted into the tissue affected. That portion of the exudate not used up in cell formation is resorbed. An example of this is seen in healing by first intention. The small amount of fluid poured out between the edges of the wound is converted to epithelium at the surface. Beneath this it forms connective tissue in which the primitive cells are elongated into tubes and threads which fuse with similar structures from adjoining cells.

2. The amount of exudate is greater than in the preceding type but there is still no accumulation of fluid. Excessive numbers of primary cells develop and are converted into the same or different tissues. Such an occurrence is seen in instances of hypertrophy, exostosis, ankylosis of joints, closing of blood vessels by organization of the clot (blastema) and hepatization of the lung in pneumonia. This concept led Legroux in 1838 to maintain that cardiac hypertrophy always resulted from a preceding inflammation, in opposition to the view of Corvisart that it was the result of increased demand and an impaired circulation.

3. The amount of fluid is large, and only a portion is converted into primary cells, the remainder escaping on free surfaces or collecting in body cavities. The quality of the exudate in this instance also affects the outcome of the process. Thus, in a thin watery exudate as seen in inflammation of a serous membrane (coryza) the number of primary cells formed is small. This is also seen on cutaneous surfaces, where the scarcity of cells is associated with slow heal-

21. Henle, J.: (*a*) J. d. pract. Heilk. **86**:3, 1838; (*b*) Arch. f. Anat. u. Physiol. **6**:1, 1839.

ing or absence of healing, producing an ulcer. On the other hand, a thick creamy pus contains many primary cells, healing is facilitated, and the term "laudable pus" is justified.

Within a year of its announcement by Schwann the cell concept as he understood it had been used as the basis for an interpretation of the two great fields of pathologic investigation —inflammation and neoplasia. Though it shed light on many phenomena previously wholly unclear, the mistaken belief in the spontaneous generation of cells within a structureless blastema led to many erroneous conclusions.

It is worth noting that the blastema as applied in pathology was an exudate and therefore derived from the blood. Investigators had been familiar with the idea of the blood as the source of organized tissues since the days of John Hunter. According to the latter, the blood was in itself alive, and this quality was concentrated in the coagulable or plastic lymph. Hunter [22] wrote:

. . . I had long suspected that the principle of life was not wholly confined to animals or animal substances endowed with visible organization and spontaneous motion: I conceived that the same principle existed in animal substances devoid of apparent organization and motion . . .

Elsewhere he said:

. . . For this [coagulation] it [blood] requires rest, either by extravasation, or by being retained in the vessels till the utility of circulation is lost. . . . Under any of these circumstances it becomes a solid body; for the moment it is at rest it begins to form itself into a solid, and changes into this or that particular kind of substance according to the stimulus of the surrounding parts which excites this coagulum into action, and makes it form within itself blood, vessels, nerves, etc.

Again, Hunter wrote:

. . . This blood so extravasated, forms either vessels in itself, or vessels shoot out from the original surface of contact into it. . . . I have reason to believe, however, that the coagulum has the power, under necessary circumstances, to form vessels in, and of itself . . .

Over fifty years later Virchow,[23] in stating his belief that angiosarcoma arises de novo from a thrombus in a vein, wrote: "Here, then, there apparently occurs the formation of a cancer from a fibrinous clot in the manner described by Hunter. . . ."

FURTHER DEVELOPMENT OF THE IDEAS OF CELL ORIGIN

If blood and plasma clots acted as the source of cells in the living body, it seemed reasonable to suppose that they might act similarly when removed from the body and treated with various chemicals. This opened a phase of study which was energetically pursued by several investigators. One of the most prominent in this field was Ascherson who drew the following conclusions in an article [24] published in 1840:

1. If albumin is contacted by liquid fat, a tough elastic membrane surrounding the albumin is always formed. 2. The tissues of animals consist of cells that may be regarded as metamorphosed oil droplets. The erythrocytes are cells that contain liquid fat and it is their chief function to carry the latter everywhere that new cell formation takes place. The primitive state of the animal ovum is that of a fat droplet.

The whole theory is similar to the earlier one of Raspail, already cited.[8]

The famous Scottish physician, John Hughes Bennett, who in 1845 was the first to describe a case of leukemia, was an adherent of Ascherson's views. He wrote: [25]

. . . Indeed, it appears to me in the highest degree probable that all blastemata containing the necessary nutritive principles in solution, precipitate minute oil particles which are the elementary granules of histologists. These, either separately or united, constitute nuclei composed of oil, surrounded by an albuminous membrane. . . . It must always be remembered that the granules produced mechanically by the union of oil and albumin are not vital structures; but where formed in the animal body, under certain conditions, they become so. . . . These considerations lead to a generalization which appears to me to be of great importance; namely, that the molecular element is the real basis of all the tissues, and not the cell, as maintained by Schwann, or the nucleus as contended by Henle.

How many have been the "ultimate" units of life! First the cell, then the nucleus, the centrosome, the chromosomes, the mitochondria—all have been pressed into service. With the advent of the electron microscope it is certain that yet another will be added to the list. The physiochemical concept of cell formation was upheld, in part at least, as late as 1852 by Panum: [26]

. . . It must be granted that in the living organism such a purely physical or chemical cell formation is conceivable alongside the true, physiologic structures capable of further development. At least as concerns the so-called fat cells it would seem unnecessary to call on incomprehensible vital powers for aid since one can artificially produce structures indistinguishable from those arising in the organism.

Yet withal progress was being made and the views hitherto expressed did not impress the

22. Hunter, J.: A Treatise on the Blood, Inflammation, and Gunshot Wounds, London, G. Nicoll, 1794.

23. Virchow, R.: Virchows Arch. f. path. Anat. (*a*) **1**:112, 1847; (*b*) **1**:201, 1847; (*c*) **1**:118, 1847.

24. Ascherson, F. M.: Arch. f. Anat. u. Physiol. **7**:44, 1840.

25. Bennett, J. H.: On Cancerous and Cancroid Growths, Edinburgh, Sutherland & Knox, 1849.

26. Panum, P.: Virchows Arch. f. path. Anat. **4**: 155, 1852.

majority of investigators. Henle, writing at the time, said:

> . . . Discovery follows discovery with such rapidity that now the enthusiasm for observation is so great that time and breath fail for the setting up of systems. May it remain so yet awhile.

As might be expected, the more rigid concepts of cell lineage were introduced by embryologists. Even before Schwann had thought of a cell, the great Karl Ernst von Baer wrote a paper [27] on the cleavage of the frog's egg. In this he pointed out that Prevost and Dumas, who had first described the phenomenon, noted only the appearance of surface furrows, not realizing that these passed entirely through the egg. Baer showed that "grooves visible at the surface are merely the boundaries of fissures that divide the entire yolk sphere." In detail he followed the individual lines of cleavage until at one stage "there are 256 corpuscles. . . . Cleavage continues until the countless little new individualities have no significance and appear only as cleavage particles of a new individual."

In 1844 Kölliker,[28a] writing on the embryology of cephalopods, said:

> . . . While investigating the structural relationships of embryos and adult animals I have arrived at the conclusion that the simple and yet unspecialized elements are derived from one another in unbroken succession, the later ones always being descended from the earlier. All segmentation spheres (blastulae) with their cells, are derived from the primary embryonic cell, its nucleus and surrounding yolk. The cells of the segmentation sphere in turn form secondary cleavage cells which pass directly into all the various tissues. Finally, the growth of the tissues in the incompletely developed animal, in so far as it is the result of cells and their multiplication, is most probably a derivative of the cleavage cells.

Five years later, in 1849, the man who wrote the foregoing passage welcomed Rudolf Virchow as a colleague to the University of Würzburg. While still at this university in 1855 Virchow was to write the famous aphorism: "Omnis cellula e cellula." As an old man Kölliker [28b] said:

> . . . In 1851 Virchow still held fast to the schwannian theory of free cell formation. It is therefore understandable that I became doubtful of the law discovered by myself in normal development. In the first edition of my "Manual of Human Histology" I accepted a free cell formation for certain normal elements. . . .

In the same year that Kölliker wrote of cephalopods, a botanist, Unger,[29] attacked the theory of Schleiden with the words:

> . . . My chief argument against this theory is that the formation of cell vesicles about the nucleus cannot be observed . . . much less are they seen to enlarge and form cells. In fact, I am not exaggerating when I maintain that no plant anatomist has ever witnessed this process in such a way as to be convinced of its existence.

Unger called attention to many delicate cross walls in growing plant tissues and believed that these were the early stages of reduplication. The frequent absence of the nucleus at this period of cell division he unfortunately interpreted as evidence that the nucleus is quite unimportant in cell multiplication.

Although his explanation was incorrect, Unger was one of the first who noticed the disappearance of the nucleus as a formed body during cell division. Had the available technic been more satisfactory, he might then and there have discovered in mitosis the most common method of cell duplication. This needed to wait until 1873 when Schneider described it in the egg cells of a platyhelminth. Until then the theory of cell division most commonly held after the overthrow of free cell formation was that of amitosis. This was first positively described by Remak in 1841, who observed it in the white blood corpuscles of tadpoles.

At this time John Goodsir wrote the "Centers of Nutrition",[30] which foreshadowed much of Virchow's work and is recognized by the latter in his dedication of the English edition of "Die Cellularpathologie" to the great Scotsman. Goodsir was aware of the functional significance of cells and cell groups as well as of their descent from preexisting cells when he wrote:

> . . . A nutritive center, anatomically considered, is merely a cell, the nucleus of which is the permanent source of successive broods of young cells which from time to time fill the cavity of their parent and pass out in certain directions and under various forms, according to the tissue or organ of which their parent is a part. . . . It would appear that from a central cell all the other cells of its department derive their origin. It is the mother of all those within its own territory.

Clearly, here is a statement of the derivation of cells from preexisting cells, even though based on the then widely accepted and erroneous concept of their endogenous formation.

Though by 1845 the cellular character of the parenchymatous organs was recognized, this did

27. von Baer, K. E.: Arch. f. Anat. u. Physiol. **1**: 481, 1834.

28. von Kölliker, A.: (*a*) Entwickelungsgeschichte der Cephalopoden, Zurich, Meyer & Zeller, 1844; (*b*) Erinnerungen aus meinem Leben, Leipzig, W. Engelmann, 1899.

29. Unger, F.: Bot. Ztg. **2**:489, 1844.

30. Goodsir, J.: Centers of Nutrition, in Goodsir, J., and Goodsir, H. D. S.: Anatomical and Pathological Observations, Edinburgh, M. Macphail, 1845; reprinted, The Anatomical Memoirs of John Goodsir, edited by W. Turner, Edinburgh, A. & C. Black, 1868.

not extend to the connective tissue. To Virchow goes most of the credit for clearing up this problem, which for a decade divided histologists into opposing camps, of which he and Henle were the respective leaders.

Henle derived elastic fibers from naked elongated nuclei, while Virchow contended that they arose from true cells. Reichert had shown that common connective tissue consisted at first only of cells between which the collagen was deposited. This view was accepted by Virchow but opposed by Henle, who claimed that what the former called cell bodies were empty spaces. Of this Virchow said: [31]

. . . I showed that in many places where it was believed that only nuclei existed, true cells were present, and that much taken to be lacunas and spaces were actually cell outlines. Similarly I have shown that there are complete cells present within the spaces in bone and cartilage. . . . The connective tissue too consists of a ground substance in which are lacunas similar to those in bone and cartilage, and these also contain cells.

These studies on the nature of connective tissue were of great importance in leading Virchow to the denial of a spontaneous generation of cells within a blastema. This was accomplished in great measure by the discovery that the organization of a fibrinous exudate could be traced to an ingrowth of fibroblasts from the surrounding connective tissue. Until the cellular character of the latter tissue had been established, the cells appearing in an organizing exudate could be accounted for only on the basis of their spontaneous appearance within the exudate. But Virchow went too far when he tried to derive most neoplasms from the connective tissue and denied that carcinoma was the result of epithelial cell proliferation. For him and the majority of other investigators during the middle of the last century, metaplasia of connective tissue was almost unlimited. Hence Virchow wrote: [32a]

. . . With little modification, one can replace the plastic lymph, the earlier blastema and the later exudate with the connective tissue and its equivalents as the common germinal tissue of the body from which the development of new parts can be traced.

Following extensive embryologic researches, Remak [33] in 1852 was able to anticipate Virchow's derivation of all cells from other cells and to foresee its pathologic implications:

. . . These results are applicable to pathology quite as much as to physiology. It can hardly be denied that pathologic tissues are only variants of the normal embryonic developmental types and it is not likely that they retain the "privilege" of extracellular cell formation. The so-called "organization of plastic exudates" and the earliest development of tumors requires proof. Supported by the proof which has now been given to my skepticism of many years' standing, I shall risk the pronouncement that the pathologic tissues are no more formed in an extracellular cytoblastema than are normal structures, but rather that they are derivatives of normal tissues of the organism.

31. Virchow, R.: Virchows Arch. f. path. Anat. **5**: 590, 1852.

32. Virchow, R.: (*a*) Virchows Arch. f. path. Anat. **8**:23, 1854; (*b*) **8**:8, 1854.

33. Remak, R.: Arch. f. Anat. u. Physiol. **19**:47, 1852.

INFLAMMATION

The study of inflammation made some progress during the years 1845 to 1858, but the best work, that of Addison and Zimmermann, remained without influence. The Vienna school, dominated by Rokitansky, taught that inflammation consisted primarily of an exudation of fluid from the capillaries. Virchow, ever more impressed with the significance of the cell, opposed this view and found in the avascular cornea a good experimental object. Under his direction, Wilhelm His, who subsequently became famous as an embryologist, wrote a thesis on the normal and the pathologic histology of the cornea.[34] Using the rabbit's eye as test object and silver nitrate to cauterize the cornea, he noted a marked increase in the number of nuclei seen in the affected part. Unaware of the migration of leukocytes, he interpreted his results as follows:

. . . It is thus apparent by simple anatomic inspection that the most intense nuclear proliferation occurs in the vicinity of the irritated area, remote from the blood vessels; whereas the marked increase in cell size reaches its height near the vessels. Between these is a region in which nuclear division as well as cellular enlargement is relatively moderate. In other words, nuclear division is a function of the cell; it is the latter's reaction to noxious agents, quite independent of any vascular influence. On the other hand, the enlargement of the cell is dependent on the materials supplied by neighboring vessels.

With these words the student has epitomized his teacher's views on the subject much as they were to be presented in "Die Cellularpathologie" under the name "parenchymatous inflammation." The increase in size and the altered character of the cells are recognized today in the vague term "cloudy swelling."

In 1843 William Addison placed a frog's foot in hot water and then observed it under the microscope. He said: [35]

. . . But it may be urged as an objection to this theory of nutrition [inflammation], that as we can see the colorless corpuscles in the irritated web of a frog's foot adhering to the tissue [endothelium], why do we

34. His, W.: Beiträge zur normalen und pathologischen Histologie der Cornea, Basel, Schweighauser, 1856.

35. Addison, W.: The Actual Process of Nutrition in the Living Structure Demonstrated by the Microscope, London, J. Churchill, 1843.

not see them passing through it? . . . To this I answer that the nutritive changes or processes are too slow in this example for us to follow from beginning to end all the actual stages of nutrition; the corpuscles go on congregating in the irritated tissue for an hour or two.

Yet in 1849 Hassall wrote: [36]

. . . With respect to the nature and origin of the granular corpuscles, some physiologists will have it that they are white corpuscles escaped from the vessels—an opinion completely untenable. They doubtless have an origin external to the blood vessels, and are to be regarded as of an epithelial character.

In that same year Addison wrote another article,[37] illustrated with figures, in which he fully anticipated Cohnheim. In his famous paper of 1867 the latter,[38] in a footnote, quoted Addison in extenso and pointed out that his attention had been called to that work by Virchow. However, in "Die Cellularpathologie" Virchow wrote:

. . . The individual white blood corpuscles have the characteristics of pus cells, and you can see that one could hold the leukocytes to be pus corpuscles, not only in the healthy person, but even more in the pathologic state when the blood or other parts are full of these elements. You can readily see how one can arrive at the question which has been seriously raised, whether pus cells are not simply extravasated leukocytes; or the converse, whether leukocytes found within the vessels are not pus cells taken up from the tissues. But a pus cell can be distinguished from a white blood corpuscle only by the manner of its origin. . . . If you do not know whence it has come you cannot say what it is.

By 1858 most investigators had abandoned the idea that pus cells appeared spontaneously in an exudate. The generally accepted belief was that the cells could arise either from connective tissue or from epithelial cells. That they were identical with leukocytes was accepted by few. In vain had Zimmermann written: [39]

. . . Inflammation can only be that local process in which an anomaly of the circulation can be demonstrated—an abnormal escape of blood corpuscles from the vessels, associated with a focal rise in temperature.

NEOPLASIA

The study of tumors was prosecuted with vigor in the years 1845 to 1858. During most of this period the cancer cells were thought to arise in a blastema, subsequent multiplication occurring by endogenous proliferation. In 1847 Virchow [23b] gave the following summary of the development of a cancerous neoplasm:

. . . As a result of changes in nutrition a gelatinous exudate occurs at a given point. This exudate is of unknown chemical composition and varies greatly in the amount of moisture it contains. Occasionally it persists as such, when it forms the gelatinous cancer. Usually, however, cells develop within it. These differentiate in two directions, becoming either connective tissue cells or cells which subsequently can no longer become connective tissue (tumor cells). Along with the connective tissue there are formed blood vessels and elastic fibers; rarely, it becomes ossified. Depending on the prevalence of this or that formation, there results a fibrous, cellular or vascular cancer. If the cells become filled with pigment a pigmented cancer develops, if extravasations alter its character we have a hemorrhagic cancer . . .

At this time Virchow was still a firm believer in free cell formation within a blastema. In defense of this view he wrote: [23c]

. . . There are cancers—and most cellular cancers belong to this group—in which no primary exudate can be found. What does that prove? Has any one seen the exudate from which arises the pus of a hepatic abscess, or that which develops into connective tissue and by its contraction leads to the granular appearance of the liver? There are processes confined to such small areas, where development proceeds so rapidly, that we are seldom able to observe the primary exudate. In such cases it suffices to apply general laws which have been discovered by other means.

Seven years later Virchow had advanced far beyond this view but was still not wholly free of it: [40]

. . . A large part, yes, perhaps the majority of new formation is the result of a progressive development of young elements from preexisting tissue cells.

During the following year he finally cast aside the idea of a spontaneous generation of cells. It was then that his famous aphorism appeared for the first time, and specifically with respect to tumors: [32a]

. . . I formulate the doctrine of pathologic generation of neoplasm in the sense of cellular pathology simply as: "Omnis cellula e cellula." [41]

36. Hassall, A. H.: The Microscopic Anatomy of the Human Body in Health and Disease, New York, Pratt, Woodford & Co., 1851.

37. Addison, W.: On Healthy and Diseased Structure, London, J. Churchill, 1849.

38. Cohnheim, J.: Virchows Arch. f. path. Anat. **40**:1, 1867.

39. Zimmermann, G.: Med. Ztg. **21**:64, 1852.

40. Virchow, R.: Handbuch der speciellen Pathologie und Therapie, Erlangen, F. Enke, 1854, vol. 1.

41. Charles Oberling (Le problème du cancer, Montreal, Les Editions de L'Arbre, 1942, p. 31) writes: "On aboutit ainsi au dogme *omnis cellula e cellula* énonce pour la première fois par Leydig et reproduit par Virchow dans un article paru en 1855." Apparently this is based on a misinterpretation of a statement of Marc Klein appearing in the latter's "Histoire des origines de la théorie cellulaire" (Paris, Hermann, 1936, p. 62). He said clearly that the aphorism was first published by Virchow in 1855. Any implication that the phrase, though not published, was first enunciated by Leydig is not supported by available evidence.

It is of interest to note at this point that Virchow did not reach this conclusion as a result of his investigations on tumors:[1b]

. . . My first experience, on the basis of which I began to doubt the dominant doctrine of blastema and exudate as a source of new formations, dates from my studies on tubercles. For I found that in a series of tubercles in various organs, particularly in the lymph nodes, meninges and lungs, at no time was there a recognizable exudate. Rather, they presented organized elements at every stage in their development; at no time was an amorphous structureless state found.

During this period Virchow's belief in the derivation of almost all tumors from connective tissue gained many adherents. His insistence thereon led to the opinion that cancerous tumors of the epidermis, which were obviously derived from epithelium, were not true cancers; they were known as cancroids. Although Virchow did not believe in a specific cancer cell common to all tumors, his restriction of the site of neoplasia to the connective tissue encouraged those who sought such a cell. The confusion which these two misconceptions wrought is well illustrated by the work of Hannover,[42] who wrote an entire book on cancer of the skin. Being convinced that the lesion arose only by a transformation of epithelial cells and lacked the specific cell he thought essential to the diagnosis of cancer, Hannover sought to solve the dilemma by coining a new term, "epithelioma." The word is in use today and to some extent still carries with it the vagueness it had at birth.

The most prominent champion of a characteristic cancer cell was Lebert, who developed his views at great length in numerous books and articles.[43] One of his colleagues in Paris was the famous surgeon Alfred Armand Velpeau, who frequently conferred with Lebert on interesting cases. One such instance was that of a testicular tumor in a young man of 18 years. Of the surgical specimen Lebert[44] wrote to Velpeau:

. . . Pressure of the tumor does not anywhere produce a juice resembling the cancerous, and on scraping it with a scalpel, nothing appears but a transparent liquid, like synovia. Examining the tissue of this tumor under the microscope it is easy to be satisfied that it is merely an ordinary fibroplastic tumor, and one even of the best conditioned [benign].

Later, when the patient was obviously cachetic, Lebert said:

. . . The microscope did not enable me to discover the cancerous cellule, but only fibers, narrow fusiform bodies, and fibro-plastic globules. I maintain then, that the tumor had not the characteristics of cancer.

He accounted for the condition of the patient on the basis of true cancers developing independently in the abdomen. Finally Lebert wrote Velpeau:

. . . I assisted this morning at the autopsy of the young man afflicted with abdominal tumors, consecutive to an operation for sarcocoele. It is certain that the rapid progress of the disease—nine months in all from its commencement—, the rapid development of the tumors in the abdomen since the operation, finally, the general cachectic state, militate in favor of the diagnosis made by you. Nevertheless, the microscopic examination of many morsels taken at the autopsy shows no globules characteristic of cancer, but simply those of the fibroplastic tissue, as in the testicular tumor you removed from this young man. . . . It appears true that one may view this case as a general diathesis of fibroplastic tumors, having a progress analogous to cancer.

Small wonder that Velpeau[44] wrote:

. . . M. Lebert and after him M. Robin, have the misfortune, in my opinion, to set down as a fact what is always a question; namely, that a certain determinate cellule forms the element of each kind of tumor. . . . To assert that cancer of the lips, face, arms, uterus, penis, integuments in general, are merely hypertrophied follicles or masses of epithelium; that warts, corns, horny productions, and steatomas are identical with the former, must appear, even *a priori*, always strange to experienced surgeons.

Virchow too was fully aware of this rift between microscopists and clinicians, for he said:[32b]

. . . I agree with Velpeau that it is in no wise necessary to call in the aid of the microscope every time in order to recognize this or that tumor. I too believe that a trustworthy diagnosis of most visible tumors can be made without a microscopic examination.

How keenly Virchow felt the ridicule to which histopathologists were exposing themselves by their insistence on the specificity of the cancer cell is apparent in one of his early papers,[45] wherein he says:

. . . The significance of microscopic observations for the theoretical problems of physicians has never been really demonstrated, and in practical matters, especially diagnosis, the greatest errors have been made by the microscopists. It need, therefore, hardly be cause for wonder that their influence upon the practice (of medicine) has been small. The microscopists have, however, been permitted to disembowel one another in arguments about this or that type of cell before the eyes of the clinical notables. The clinicians were amused by corpuscles with tails, and wondered why they didn't also possess scissors. They sat politely smiling upon

42. Hannover, A.: Das Epithelioma, eine eigenthümliche Geschwulst, die man im Allgemeinen bisher als Krebs angesehen hat, Leipzig, L. Voss, 1852.

43. Lebert, H.: Physiologie pathologique, Paris, J.-B. Baillière, 1845.

44. Velpeau, A.: A Treatise on Cancer of the Breast, translated from French by W. Marsden, London, Sydenham Society, 1856.

45. Virchow, R.: Virchows Arch. f. path. Anat. **1**: 207, 1847.

the dais while the barbarians were slaughtering one another . . . Occasionally one heard a young clinician say with a disparaging gesture, "Oh, that's probably histological!"

CONCLUSION

It was this skepticism on the part of the clinician which needed to be overcome. If the new achievements in embryology, histology and pathology were to gain wider acceptance, if they were to become a part of the physician's scientific armamentarium, it was necessary that they be carried from the study and the laboratory into the great lecture halls and clinics.

For this task Virchow was eminently suited. As an outstanding contributor to original work in the fields of normal and pathologic histology he could speak with authority. As a physician he knew the requirements of his colleagues, and as professor at the University of Berlin he had their respect. In the spring of 1858 he gave a series of lectures to physicians and graduate students of medicine. They were intended to serve as an introduction to the existing knowledge concerning the cellular structure of the body and its application to pathology and medicine.

As published under the title "Die Cellularpathologie," these lectures contained nothing that was new and much that subsequent work proved to be wrong. But though this book added little to pathology, it made the physician aware of what pathologists had accomplished and what their findings meant to him. For this reason most physicians of today look on "Die Cellularpathologie" as the primary source of the concept for which it is named. That it is something less than this does not detract from its greatness. The purpose of this article has not been to remove it from the ranks of the great masterpieces, but rather to show why it has been placed there.

Books Received

VIRUS DISEASES IN MAN, ANIMAL AND PLANT. By Gustav Seiffert. Pp. 332. Price $5. New York: Philosophical Library, 1944.

TUBERCULOSIS OF THE EAR, NOSE AND THROAT. By Mervin C. Myerson, M.D., New York. Pp. 291, with 88 illustrations. Price $5.50. Springfield, Ill.: Charles C Thomas, Publisher, 1944.

MANUAL OF HUMAN PROTOZOA. By Richard R. Kudo, D.Sc., associate professor of zoology, University of Illinois. Pp. 125, with 29 illustrations. Price $2. Springfield, Ill.: Charles C Thomas, Publisher, 1944.

THE MANAGEMENT OF NEUROSYPHILIS. By Bernhard Dattner, M.D., Jur.D., associate clinical professor of neurology, New York University Medical College, with the collaboration of Evan W. Thomas, M.D., assistant professor of medicine and assistant professor of dermatology and syphilology, and Gertrude Wexler, M.D., instructor in dermatology and syphilology, New York University Medical College. Pp. 398, with 7 illustrations. Price $5.50. New York: Grune & Stratton, 1944.

INFECTIONS OF THE PERITONEUM. By Bernhard Steinberg, M.D., director of Toledo Hospital Institute of Medical Research; past fellow of the National Research Council; former Crile Research Fellow, Western Reserve University. Pp. 455, with 45 illustrations. Price $8. New York and London: Paul B. Hoeber, Inc., 1944.

This book deals with the causation, the genesis, the course, the diagnosis and the treatment of peritonitis. It is written in the light of a thorough understanding of the basic problems. The author has correlated well with current knowledge and conceptions the results of his own work, experimental and observational, on peritonitis during the past eighteen years. The book is an important contribution toward the advancement of the control and treatment of peritonitis. It will be of interest and instruction to all who are concerned with the practical and scientific problems of peritoneal infections.

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